THE PHILIPPINE

JOURNAL OF SCIENCE

VOL. I FEBRUARY 15, 1906 No. 2

THE KEEPING QUALITIES AND THE CAUSES OF RANCIDITY IN COCONUT OIL.

By HERBERT S. WALKER.

(From the Chemical Laboratory, Bureau of Science.)

In almost every work on fats and oils, coconut oil is cited as being especially prone to become rancid. Lewkowitsch[1] states that, when fresh, it possesses a bland, pleasant taste and odor, but that on standing it quickly becomes rancid. Samples analyzed by him contained from 5 to 25 per cent of free acid. Schestakoff[2] says that pure coconut oil shows an acid value (milligrams of caustic potash) of from 2 to 5. On standing under abnormal conditions, this may in one year rise to 60 or 70.

Coconut oil is in enormous demand as the basis of edible products such as "vegetable butter," etc., and therefore it is of the utmost importance to be able to produce an oil which, as nearly as possible, is free from fatty acids, rancid odor or taste, and which at the same time may be shipped without fear of deterioration.

The experiments to be described were undertaken with the view of discovering the conditions which induce a rapid deterioration of coconut oil, and, if possible, of ascertaining a means of improving its keeping qualities. In the course of this work it was noticed that the oil does not change with as great rapidity as is generally believed to be the case. The ordinary commercial oil, bought in Manila, contains from 5 to 10

[1] Lewkowitsch: *Chemical Analysis of Oils, Fats, and Waxes.*

[2] Schestakoff: Über den Gehalt an freien Fettsäuren natürlicher Fette und Öle. *Chem. Rev. Fett. u. Harz. Ind.* **9**, 180.

per cent free acid, and there is no very great increase in acidity even on prolonged standing. Mr. Richmond, of this laboratory, in some experiments on a commercial oil, showed that it was not affected by passing a current either of dry or of moist air through the liquid for five days. An oil prepared by drying fresh coconut meat at 80° to 90° C., and then extracting with petroleum ether, was sterilized and sealed in a glass tube on August 16, 1904. At that time it contained 0.13 per cent free acid (calculated as oleïc). This sample was allowed to stand until February 21, 1905, and it was then again tested, with the result that it was in as good condition as at the time of the previous examination; there had been absolutely no increase in free acid. A sample of the same oil, kept in a sealed tin tube, on February 21, 1905, contained 0.34 per cent of free acid, an increase of only 0.21 per cent in six months. This same oil was allowed to remain in tin, unsealed, until April 11, 1905; the acidity had then increased to 0.46 per cent. On further standing in a glass-stoppered bottle until August 16, 1905, the figure was 0.50 per cent. An expressed oil from the same preparation of copra, kept under similar conditions, showed:

	Per cent.
February 21	0.77
April 11	0.78
August 16	0.96

This oil, from the start, had a slightly burnt odor and taste and in time it deposited a dark-brown sediment. However, neither of the two oils showed any signs of "rancidity" and even after a year had elapsed they were almost as pleasant to the taste as when first prepared.

In order more fully to study the effect of age and method of preparation on the keeping qualities of coconut oil, the following samples were prepared, their condition noted, and their exact titer determined. These oils will be allowed to stand for several years if necessary, until final results are obtained as to their respective rates of deterioration, but in the meanwhile the change up to the present time is given in the table which follows:

DESCRIPTION OF OILS USED AND DESCRIBED IN TABLE I.

(A) Expressed oil from vacuum-dried copra. Has been heated for two hours at 100° and filtered twice through paper. A light-colored, clear oil with the characteristic coconut taste and odor.

(B) An oil similar in every respect to "A" except that it was prepared from copra dried at 80° to 90°, without vacuum.

(1) Fresh coconut meat grated and dried at 80° to 90° on August 16, 1904; was allowed to stand in a covered specimen jar until March 11, 1905. At that time it was still of a pleasant odor and taste, although both odor and taste were not quite as good as when the specimen was freshly prepared. No mold growth was present. A sample of oil was expressed from a portion of this copra by using a hydraulic press with a final pressure of 450 kilograms per square cen-

timeter. This oil, after filtration, was of a light-yellow color and it was of a pleasant, although slightly burnt, odor and taste.

(2) Oil No. 1 was heated at 100° for three hours, while at the same time a current of air in a partial vacuum was passed through it. This process leaves the color and free acid unchanged but removes almost all of the burnt odor, leaving a bland, almost tasteless, oil.

(3) An oil from the same copra as Nos. 1 and 2 but prepared by extraction with petroleum ether. Afterwards it was treated in the same manner as No. 2. It differs from Nos. 1 and 2 in being practically colorless.

(4) Commercial coconut oil treated with alcohol and animal charcoal and then filtered; the alcohol was afterwards distilled and recovered. This oil was rather unpleasant to the taste, but it had no odor.

(5) Commercial coconut oil treated with live steam; this removes the odor, but the unpleasant taste remains.

(6) Fresh meat, ground and dried in vacuum at 70° to 80°. The oil was expressed and once filtered; it possessed a very pleasant coconut-like odor and taste. It still contained a considerable amount of sediment.

(7) Coconuts cut in halves and dried in vacuum at 75° to 85°. The oil expressed and filtered twice. It had a very pleasant odor and taste.

(8) The same oil as No. 7, heated at 100° for one and one-half hours and filtered hot.

(9) The same as No. 7, heated at 100° for one and one-half hours, while at the same time a current of air was passed through the oil under partial vacuum. Filtered hot and bottled.

(10) Fresh coconut meat, ground and pressed in a hand press to remove most of the milk. Afterwards this meat was dried completely by spreading it in the sun for about five hours. The oil expressed from this copra was almost water white and without taste or odor.

(11) Coconuts split in halves and dried in the sun for five days. Ground and expressed. Yielded a cloudy, light-colored oil, very hard to filter, with a peculiar but not unpleasant taste and odor. This sample was strained through cloth but not filtered.

(12) Same as No. 11, strained and filtered slowly through paper.

(13) Same as No. 11, heated at 100° for two hours and filtered through paper.

(14) Fresh nuts, split in halves and allowed to stand during one week in the air at room temperature (about 30°). A vigorous mold growth and an unpleasant odor developed. This moldy meat was dried in a vacuum and the oil was expressed. This was highly colored and was rather unpleasant to taste and smell.

(15) Commercial coconut oil shaken with 2 per cent of solid calcium oxide (burned lime), heated to 100° and filtered. The filtrate was treated with animal charcoal and again filtered; there resulted a colorless oil which was very free from an unpleasant odor or taste.

(16) The same copra as that used for No. 1; was allowed to stand one month longer in an open jar, then expressed.

(17) Oil expressed from vacuum-dried copra which had stood for one month exposed to the air; the oil was heated to 100° and filtered.

(18) Expressed from sun-dried copra and treated in the same manner as No. 17. Both of these samples were of as pleasant a taste as oils from fresh copra.

(19) Vacuum-dried copra which had stood in a closed desiccator over water for one month, and which had accumulated a very decided growth of mold. It was dried for one hour and expressed. The oil had a considerable color and was slightly unpleasant as to taste and odor. Heated to 100° and filtered.

(20) Sun-dried copra treated in the same way as No. 19. Yielded an oil somewhat darker in color but otherwise much the same as No. 19. Filtered without heat.

(21) Same as No. 20, heated to 100° before filtering.

(22) The same copra as that used for samples 1 and 16 was allowed to stand for three weeks over water and for one week in air, and then dried and pressed. A vigorous mold growth appeared in the copra and a peculiar ethereal odor was apparent. The oil itself was of a light-yellow color, with a pungent, rather unpleasant, odor and an extremely disagreeable taste.

(23) Expressed from commercial copra, first quality, sun dried, Tacloban, Leyte. The unfiltered oil is dark colored and cloudy, depositing a black sediment.

(24) Same as No. 23, filtered. Almost colorless.

(25) Expressed from commercial copra, grill dried, Laguna (second quality). Not filtered.

(26) Same as No. 25, filtered. Light yellow in color.

(27) Expressed from commercial copra, grill dried, Romblon (considered second quality). The filtered oil is light yellow in color.

(28) Expressed from commercial copra, first quality, sun dried, Iloilo. The filtered oil is light yellow in color.[8]

(29) "*Langis*" coconut oil, prepared by the customary native process of grating the fresh meat, exhausting it repeatedly with water, and boiling down the emulsion thus obtained until it is nearly dry. The oil is then poured off from the brown coagulum which sinks to the bottom of the vessel. A freshly prepared oil, isolated in this manner, is very light in color and it possesses a decidedly pleasant coconut odor and taste. Before filtration it is more or less turbid, owing to the presence of a small amount of water and of albuminoids.

(30) Same as No. 29, filtered. The oil is water white.

(31) Best grade commercial coconut oil, probably made from fresh meat. It is light colored but very turbid and contains considerable water and suspended matter.

(32) Commercial coconut oil, probably made from copra. Very clear but highly colored.

(33) Commercial coconut oil, Manila. Probably made from fresh meat. It contained considerable suspended matter and water.

(34) Commercial coconut oil, Cebu. A highly colored "rancid" oil. Considerable sediment in the bottom of the bottle.

(35) Commercial coconut oil, Tayabas. A highly colored rancid oil made from copra. It is only a few months old.

The following table shows the change in the amount of free acid which has been produced in these oils while they were standing from the time of their expression up to the date of writing. The free acid was determined in each case by dissolving a known weight (about 5 grams) of oil in 50 cubic centimeters of neutralized absolute alcohol, and

[8] The copras from which these last six samples of oils were made were secured through the courtesy of Messrs. Smith, Bell & Co. and the Compañía General de Tabacos de Filipinas, and taken directly from their warehouses. The samples obtained were the ordinary grades of copra, ready for export, and had been stored for about two months, during the dry season. The oils, while not especially unpleasant to the taste, were of a sufficiently rancid character to preclude their use as edible products unless they were first subjected to a refining process.

then titrating with aqueous $\frac{N}{10}$ potassium hydroxide, using phenolphthaleïn as an indicator:

TABLE I.—*Percentage free fatty acids (as oleïc).*

No.	At start.	Two months.	Four months.	Six months.
A[a]	0.06	0.06	0.09	0.60
B	0.06	0.06	0.08	[b]0.48
1	1.2	1.3	1.5	1.9
2	1.2	1.5	1.5	1.7
3	1.4	1.6	2.1	2.6
4	5.3		5.9	6.1
5	5.5			7.6
6	0.10	0.16	0.19	0.30
7	0.16	0.18	0.19	0.27
8	0.16	0.14	0.19	0.30
9	0.16	0.16	0.18	0.25
10	0.16	0.16	0.21	0.28
11	0.13	0.18	0.25	0.28
12	0.13	0.10	0.10	0.14
13	0.13	0.09	0.09	0.15
14	3.5	3.7	4.0	4.3
15	0.32		0.38	
16	1.6	1.7	2.0	
17	0.09	0.09	0.14	0.16
18	0.16	0.18	0.25	0.27
19	1.18	1.14	1.34	1.58
20	0.69	0.69	0.74	0.85
21	0.69	0.69	0.74	0.82
22	23.3			
23	1.4	1.6	1.8	2.0
24	1.4	1.5	1.7	1.8
25	2.6	3.4	3.6	3.9
26	2.6	2.6	3.1	3.5
27	2.1	2.4	2.5	2.8
28	3.0	3.5	4.0	4.7
29	0.08	0.38	0.60	0.69
30	0.08	0.13	0.16	0.19
31	2.0	2.9		
32	6.8	7.5	7.9	8.1
33	5.5		6.9	7.2
34	8.7		10.2	11.0
35	5.0	5.5		

[a] Through the courtesy of Prof. A. H. Gill, of the Massachusetts Institute of Technology, I have been given some further data on the keeping qualities of this oil. A sample packed in a sealed tin can was shipped to him at Boston and tested there when it was about three months old. On arrival it was described as being almost water white and of perfectly sweet odor. It then contained only 0.068 per cent free acid as oleic, a figure which corresponds very closely to that obtained here at the same time.

[b] This oil was kept in a large bottle. A sample in a small bottle showed an acidity of only 0.09 at this time.

At the present time there has been so little change in any of these samples that no very definite conclusions can be drawn as to the conditions which cause rancidity on standing. However, it may be considered as established that a pure, fresh, coconut oil can be prepared which contains a minimum amount of free acid and which shows no unpleasant

taste or odor. Such an oil very slowly increases in acidity, and, even after standing for one year under ordinary conditions, may still be edible without further purification.[4] Commercial oils, on the other hand, which contain from 5 to 10 per cent of free acid when freshly prepared, deteriorate much more rapidly, even though they have been filtered and are free from impurities. For example, No. 32, which, on first examination, had 6.8 per cent of free acid, increased in two months to 7.5 per cent. This oil is very clear, bright, and dry, being entirely free from sediment or turbidity of any kind. The samples prepared by us from fresh copra ranged from 0.06 to 0.16 per cent, the increase in two months being so small as to be almost negligible. Samples Nos. 1, 2, and 3, which contained a little over 1 per cent of free acid when fresh, increased from 0.1 to 0.3 per cent in the same time.

In fact, the increase in free acid to be expected in an oil when it is standing under ordinary conditions may almost be considered as being roughly proportional to its initial acidity. There are also indications that an oil from which albuminoids, etc., have been removed by filtration will retain its original condition better than one containing the above impurities. No. 6, for instance, which has been filtered once but which contained a considerable sediment, increased in four months from 0.10 per cent to 0.19 per cent, while Nos. 7, 8, and 9, oils prepared in a similar manner but filtered more thoroughly, only showed an increase in the same time of from 0.16 per cent to 0.19 per cent of free acid.[5] This fact was a little more noticeable among oils prepared from sun-dried copra. Samples Nos. 11, 12, and 13 were taken from the same lot of oil, the only difference being that No. 11 was left unfiltered, while the impurities were removed as completely as possible from Nos. 12 and 13. It will be noticed that No. 11, in six months, shows a total increase of 0.15 per cent free acid, having a little more than double its original acid value. Nos. 12 and 13 have in the same time increased only 0.01 and 0.02 per cent, respectively.

The oils prepared from commercial copra likewise show this distinction to a greater or less extent. No. 25, an unfiltered oil, increased from 2.6 to 3.4 per cent in two months, while No. 26, which is the same oil filtered, shows no change at all. However, contrary to expectations, the difference

[4] The sudden increase of acidity in samples "A" and "B" between the fourth and the sixth months is due to the abnormal conditions under which they were kept at this time. The two samples when originally prepared were kept in 500-cubic centimeter bottles which were nearly full, but during the fourth and fifth months they were opened so frequently for the purpose of taking samples for aldehyde and peroxide tests that only about 25 cubic centimeters remained in each bottle. The increase in acidity is probably due to a continuation of the surface oxidation which is discussed in a later part of this paper. A portion of sample "B" which had previously been removed to a smaller bottle showed practically no change at the end of six months.

[5] On further standing, however, the difference in this case is not so marked.

in behavior between a filtered and an unfiltered oil is most pronounced during the first few months. On longer standing, the acid values tend to approach each other.

The most important fact brought out by this work is that by far the greatest deterioration which an oil undergoes takes place in the copra itself. After an oil has been expressed from the dried meat, its change on standing is very slight compared with that which is found in the same time while it is in the copra. For instance, sample No. 1 was prepared from anhydrous copra, which had stood in a closed jar during about seven months (under much better conditions than copra is ordinarily kept); its free acid was 1.2 per cent; an oil expressed from this copra when it was fresh had only 0.77 per cent of free acid after it had stood for seven months in tin; this same copra after remaining three weeks over water and one week more in the air yielded an oil containing 23.3 per cent free oleic acid. Samples Nos. 19 and 20 were prepared from copra which, when fresh, gave an oil with almost no free acid. Fresh coconut meat on standing for even a short time in the air becomes covered with mold and produces an oil of a more or less rancid character (cf. No. 14). No great amount of rancidity was developed in any case until signs of mold or bacterial growth were visible on the surface of the copra. From this it would seem very probable that the splitting up of fat and the accompanying "rancidity" produced in copra are in a large measure due to the action of micro-organisms, which have an excellent culture medium in the sugar, albuminoids, and water which exist, together with the oil, in coconut meat.

Koenig, Spiechermann, and Bremer,[6] in their valuable paper on the decomposition of fats by micro-organisms, have conclusively shown that cottonseed meal containing a sufficient amount of water, is attacked by molds and bacteria, and that the oil therein is, on long standing, almost completely destroyed. In the accompanying experiments the methods used by these authors were followed, with certain modifications, which consisted chiefly in substituting freshly prepared anhydrous copra for cottonseed meal, and in paying especial attention to the amount of free acid developed.

The copra used for this work was prepared by grinding up fresh coconut meat and drying it at 90° to 100° C. under a partial vacuum, until the meat was anhydrous. It was then kept over sulphuric acid, to be used as needed. This product had become quite brown during the prolonged drying, but yielded an almost colorless oil, of a sweet taste, and which contained about 0.15 per cent free acid as oleic.

Ten-gram samples were weighed out in large, stoppered test tubes and each tube was inoculated with one drop of a solution made from some

[6] Koenig, Spiechermann, und Bremer: Beiträge zur Zersetzung der Futter- und Nahrungsmittel durch Kleinwesen. I. Die Fettverzehrenden Kleinwesen. *Ztschr. f. untersuch. d. Nahrungs-u. Genussmittel* (1901), 4, 721, 769.

old, moldy copra. A definite volume of distilled water was then added to each, and all were allowed to stand for one week at room temperature (25°–30° C.) and for the same length of time in an incubator at about 35° C., the tubes being opened every morning and any change in odor or appearance noted. After two weeks the whole series was dried, weighed, the oil extracted with chloroform, and its acidity determined.

	No. 1.	No. 2.	No. 3.	No. 4.	No. 5.	No. 6.	No. 7.	No. 8.	No. 9.
Weight of dry copra	10.00	10.00	10.00	10.00	10.00	10.00	10.00	10.00	10.00
Weight of water added	0.00	0.50	1.00	1.50	2.00	3.00	5.00	7.00	10.00
Percentage moisture	0.00	4.76	9.09	13.04	16.67	23.08	33.33	41.18	50.00
Weight of dry copra at end of experiment	10.00	9.99	9.70	9.20	9.30	9.10	9.20	8.97	9.15
Gain (+) or loss (—)	0.00	—0.01	—0.30	—0.80	—0.70	—0.90	—0.80	—1.03	—0.85
Weight of oil	6.79	6.77	6.61	6.64	6.68	6.82	6.78	6.67	6.84
Gain (+) or loss (—)	0.00	—0.02	—0.18	—0.15	—0.11	(+0.03)	—0.01	—0.12	(+0.05)
Gain (+) or loss (—) of substances other than oil	0.00	(+0.01)	—0.12	—0.65	—0.59	—0.93	—0.79	—0.91	—0.90
Percentage free acid	0.15	0.18	8.7	5.2	3.9	0.46	3.0	6.1	2.9

Nos. 1 and 2 remained unchanged in appearance and odor throughout the experiment. Nos. 3, 4, and 5 developed a slight mold growth and a peculiar ethereal odor, not especially unpleasant except in the case of No. 5. No. 6 showed no mold growth but turned much darker than the others and almost from the start possessed a very disagreeable odor. Nos. 7, 8, and 9 soon developed a sour smell, which, however, was not so unpleasant as that of No. 6, but they showed no mold until they had been placed in the incubator, when Nos. 7 and 8 became covered with a vigorous white growth with numerous patches of red.[1] No. 9 remained unchanged in appearance.

The percentage of free acid in the oil here seems very closely to follow the appearance of a visible mold growth in the copra, being at a maximum in No. 3, where the mold first makes its appearance, decreasing slowly, with added moisture, up to No. 6 (no mold growth in evidence), increasing again in Nos. 7 and 8 (reappearance of mold growth).

As might be expected in an experiment of so short a duration, the loss in total weight of oil was in no case large, but it was sufficiently marked to show that it also chiefly took place in those tubes which contained a

[1] Subsequent experiments indicate that this growth in Nos. 7 and 8 was due to a loss of moisture while in the incubator.

growth of mold; the loss of substances other than oil, on the contrary, was considerably less where the mold was most vigorous.

As nothing was known concerning the organisms with which these tubes had been inoculated, it was decided to repeat this experiment, without inoculating the tubes directly, but to start the growth by simply exposing them to the action of such organisms as might be present in the air from day to day. Therefore, the tubes were filled as before and allowed to stand at room temperature for two weeks, being opened and exposed to the air for a few minutes each day. A similar set was prepared at the same time and afterwards given to Dr. Edwards, of this Bureau, for bacteriological examination.

	No. 1.	No. 2.	No. 3.	No. 4.	No. 5.	No. 6.	No. 7.	No. 8.	No. 9.
Weight of dry copra	10.00	10.00	10.00	10.00	10.00	10.00	10.00	10.00	10.00
Weight of water added	0.00	0.50	1.00	1.50	2.00	3.00	5.00	7.00	10.00
Percentage moisture	0.00	4.76	9.09	13.04	16.67	23.08	33.33	41.18	50.00
Weight of dry copra at end of experiment	9.99	10.00	9.67	9.59	9.39	9.11	9.42	8.97	(a)
Gain (+) or loss (−)	(−0.01)	0.00	−0.33	−0.41	−0.61	−0.89	−1.58	−1.03	.....
Weight of oil	6.83	6.85	6.24	6.18	6.54	6.92	6.81	6.83	.....
Gain (+) or loss (−)	0.00	(+0.02)	−0.59	−0.65	−0.29	(+0.09)	−0.02	0.00	.....
Gain (+) or loss (−) of substances other than oil	(−0.01)	(+0.02)	+0.26	+0.24	−0.32	−0.98	−0.56	−1.03	.....
Percentage free acid	0.15	0.17	11.8	12.9	13.7	0.57	0.39	0.47	0.24

[a] This tube was broken while drying.

These tubes behaved very much like those which had been used in the previous experiment, Nos. 4 and 5 showing a growth of mold and an ethereal odor in four and No. 3 in six days; No. 6 darkened and became putrid, while Nos. 7, 8, and 9 simply turned "sour."

The bacteriological examination showed no organisms to be growing in Nos. 1 and 2; bacteria were found in Nos. 3 to 9, inclusive, and molds in Nos. 3, 4, and 5 only; the latter were much more numerous than the bacteria in Nos. 3 and 4, and about equally divided in No. 5.

The mold most commonly occurring in Nos. 3, 4, and 5 was identified as *Aspergillus flavus;* others, mostly *Aspergilli,* were also found but as yet have not been indentified. Quite a number of bacilli were isolated in pure cultures, but no attempt at identification has yet been made.[8]

[8] Experiments are now being undertaken to study the action of pure cultures of all these organisms on copra of varying degrees of moisture. The cultures are being prepared by Dr. Edwards. The results will be published in a later paper.

In this series of tubes, as well as in the preceding one, a high acid value, accompanied by a loss in weight of oil, is evident only in those samples which have been attacked by molds—that is, in Nos. 3, 4, and 5, which had a water content of from 9.09 to 16.67 per cent.

No. 6, the dark-colored sample with a very disagreeable odor, showed a slight gain in the weight of oil, probably due to the production, by organisms, of bodies other than oil which are soluble in chloroform. In all the other samples the weight of the oil practically remained unchanged. The large loss in substances other than oil (sugars, albuminoids, etc.) is confined, on the contrary, to those tubes in which bacteria predominate—that is, those containing more than 16.67 per cent of moisture—indicating that bacteria obtain their carbon and hydrogen chiefly from the sugars, albuminoids, and cellulose which are present in copra, while molds directly attack the oil. Whether molds alone can split up and assimilate oil from copra, or whether they may not be symbiotic with certain bacteria, remains to be established by means of the experiments to be undertaken with pure cultures.

The most important point to be considered from a practical point of view is the fact that copra containing as little as 9 per cent of moisture is still attacked by molds, with the consequent production of free acid and coloring matter as well as loss in weight of oil. Unfortunately, the copra produced in the Philippine Islands ordinarily contains from 9 to 12 per cent of water, a condition which is the most favorable for mold growth and for the deterioration of the oil. The remedy for this is obvious. A more complete drying, to reduce the water content to 5 per cent or less, will produce a copra which is unattacked by organisms. Such a product, kept dry, will remain fresh and sweet for a long time. In a previous part of this paper I have shown that copra, once sufficiently dried, may be kept during the dry season in Manila without any change whatsoever, but recent experiments prove this not to be the case during the rainy one, even with anhydrous copra.

Two samples of the latter, cut into fine pieces, were exposed, in open specimen jars, for a period of one month. At the end of this time one sample was covered to exclude air, while the other remained open. The covered sample soon developed a slight mold growth and a characteristic ethereal odor, and at the end of another month the oil extracted from it contained free acid to the amount of 3 per cent. The sample left uncovered for two months was not changed as much, for the oil from it contained only 0.89 free acid. This is probably due to the fact that during the time of exposure there occurred several comparatively dry periods of from three to four days each, during which there was very little rain, thus giving the specimen an opportunity to become partially dry so that the beginning growth of any mold would be stopped. The covered and uncovered samples were found to contain 7.8 and 6 per cent of moisture, respectively, which indicates the marked influence of a

comparatively small amount of water on the keeping qualities of copra. As shown in the previous experiment, copra containing 4.76 per cent of moisture remains practically unchanged on standing under conditions which preclude the absorption of water, while that with 9.09 per cent produced 11.8 per cent free acid in two weeks. Between these two extremes come the two samples mentioned above, the one with 6 per cent of water increasing to 0.89 per cent and that with 7.8 per cent rising to 3 per cent of free acid during a period of two months.

EXPERIMENTS ON COPRA DRYING.

Since the quick and thorough drying of copra has been shown to be of such vital importance in order to insure the production of a pure oil, an investigation of various methods of copra drying has been made, taking into consideration not only the processes common in these Islands but also those which are used in other countries.

Sun drying.—As has been stated in the introduction to this series of papers, the simplest and most primitive mode of drying copra is to expose the nuts, cut in halves, to the action of the sun during about five days. This method, although it is a slow one, under favorable climatic conditions produces a very fair quality of copra. However, a sudden rainstorm or a succession of cloudy days is sufficient to start mold and bacterial growth, with the consequent deterioration of the copra. Considerable loss due to the attacks of insects and animals is also suffered during the long period of drying, and the finished product very seldom contains less than 9 per cent of moisture.

Grill drying.—A much quicker method is the one carried out by laying the half nuts, face downward, on a bamboo grating placed over a slow fire of coconut husks. After being dried in this manner over night the nuts are removed from their shells and are then again placed over the fire, where they are allowed to remain for from four to five hours longer. This process, although it is cheap and comparatively rapid, has the disadvantage of yielding a dark-colored product which has a smoke-like taste and odor, and it also tends to form a hard, burnt coating over the surface of the nut while the inside is left in a comparatively moist state, a fact which is often taken advantage of by the small producers, who sell their copra by weight. Commercial copra prepared in this way contains from 9 to 13 per cent of moisture.

Hot-air drying.—This method of desiccation has been used successfully for a long time in the preparation of coffee, cacao, dried fruits, etc., and is at present in quite extensive use for the making of copra in Ceylon,[9] where it is said to give a very pure, light-colored product. The type of apparatus used in that island essentially consists of a large chamber filled with wire trays upon which the coconuts are placed and over which a

[9] *Tropical Agriculturalist*, 23, No. 10, supplement.

current of hot air, driven by a fan, is passed. In Trinidad,[10] British West Indies, there is now in operation a rotary hot-air drier which, it is stated, is better than any other apparatus now in use.

For the purpose of testing the efficiency of the stationary form of hot-air drier, a double-walled, rectangular galvanized-iron box, having an internal capacity of about 0.2 cubic meter, was constructed. Three galvanized-iron trays, perforated at one end, were set in this box in such a manner that the stream of hot air entering through a 20-centimeter pipe at the bottom was compelled to pass over each in turn before escaping at the top of the apparatus. A constant current of air was obtained by means of a small electric fan which was connected with a section of 15-centimeter pipe, so arranged that it could be heated by a small kerosene stove to any desired temperature. The apparatus had a maximum capacity of 24 nuts split in halves or 12 nuts when shredded.

Experiment I.—Four nuts were split in halves and placed on the bottom tray.

Temperature of entering air, 56° C.
Temperature of escaping air, 51° C.
Time of drying, 20 hours.

The copra dried at this comparatively low temperature was very white and of the best quality. A sample of oil expressed from it contained 0.08 per cent free acid.

Experiment II.—The meat from twelve nuts was shredded by hand and treated for one day in the same manner as in the preceding experiment; it was then allowed to stand at room temperature over night and completely dried on the following day. The substance in the bottom tray naturally desiccated much more rapidly than in the other two, therefore as soon as one tray was completely dry it was removed and replaced by the one just above it.

Temperature of entering air, 56° C.
Temperature of escaping air, 50° C.
Actual time of drying:
Top, 14½ hours.
Middle, 12½ hours.
Bottom, 9½ hours.

The less completely dried copra in the two upper trays became slightly "soured" while standing over night. This caused a slight increase in free acid as follows:

Tray.	Per cent free fatty acid.
Top	0.32
Middle	0.16
Bottom	0.13

[10] *Journal d'Agriculture Tropicale* (1904), 103.

Experiment III.—The meat from four nuts was shredded and placed in the bottom tray, being stirred every half hour.

Temperature of entering air, 93° C.
Temperature of escaping air, 74° C.
Time of drying, 3½ hours.

The copra thus produced was thoroughly dry, very white, and pleasant to the taste. The oil expressed from it contained only 0.06 per cent free fatty acid.

In Experiment II, as would naturally be expected, it is evident that the meat farthest away from the entering air requires a much longer time for drying than does that which lies closer to the bottom of the box. This is due to the fact that the air gradually becomes cooler and more completely saturated with water vapor as it passes over the moist copra. For practical use, therefore, a drier should be equipped with some sort of a mechanical carrier which would constantly introduce fresh coconut meat at the coolest part of the machine and then bring it slowly down toward the hottest portion.

Experiment IV.—This was undertaken in an endeavor to ascertain the approximate time required completely to dry the fresh meat, introducing it at the top of the apparatus and shifting it gradually toward the bottom. Four trays, each containing the freshly grated meat of 4 coconuts, were prepared, and three of these were placed in the drier simultaneously, tray No. 1 being at the bottom. After the latter had become sufficiently dry, it was removed from the apparatus and tray No. 2 moved down to take its place; this was next replaced by No. 3, and finally in the same manner by the moist sample No. 4.

Entering air, 95° C.
Escaping air, 70° C.
Actual time of drying:
No. 1, 4½ hours.
No. 2, 5½ hours.
No. 3, 6¾ hours.
No. 4, 4 hours.

From the above experiments it may be concluded that the average time of drying, where the apparatus is run continuously at 95° C., will approximately be four hours.

The rotary drier.—A section of galvanized-iron pipe 20 centimeters in diameter by 6 meters long was set up on wheels and connected with a small electric motor so that it could be made to revolve at any desired speed. The same current of hot air which was previously used for the stationary drier was connected with this apparatus. Four strips of angle iron extending throughout the length of the pipe served to keep the moist copra in constant motion during the time of drying. After much preliminary work to determine the proper inclination necessary to allow the material to pass through the apparatus with sufficient slowness, it was

found that by careful manipulation the grated meat from four nuts could be dried in about two hours so as not to contain more than 6 per cent of moisture. The only objection to this method consists in the difficulty of regulating the speed with which the ground meat passes from one end of the apparatus to the other. This is dependent on four factors: (1) The number of revolutions per minute, (2) the angle of inclination, (3) the specific gravity of the coconut meat, and (4) the speed of the entering current of hot air. In the machine used here an unfortunate tendency toward a separation of the moist from the dry copra appeared; the dry particles, being lighter, were held back by the current of air or even blown out through the upper end of the tube, whereas the moist and consequently heavier pieces passed through too quickly. When these mechanical difficulties are solved this should prove the ideal method for drying coconut meat for oil-making purposes.

Vacuum drying.—The apparatus used was a small, barrel-shaped iron chamber, about 34 centimeters in diameter and in length, insulated with asbestos and heated by three hollow steam plates upon which the substance to be dried was placed. The pump connected with this drier gave a vacuum of about 660 millimeters (absolute pressure of 100 millimeters).

Experiment I.—Four coconuts (the maximum capacity of the apparatus) were split in halves, after removing the outer husk, and kept in the drier for three hours. The meat had then contracted sufficiently to allow of its being removed from the shell. During this time the temperature had gradually risen from 30° to 80°. The meat was then subjected to a further drying during four hours, at the end of which time, though not perfectly anhydrous, it was fully as dry as the ordinary commercial article.

Actual time of drying, 7 hours.
Maximum temperature, 80° C.
Vacuum, 635 millimeters.
Steam pressure, about 0.7 kilo per square centimeter (10 pounds).

Experiment II.—The preceding experiment was repeated under practically the same conditions, except that the nuts were allowed to dry completely without removing the shell.

Time of actual drying, 8 hours.
Maximum temperature, 80° C.
Vacuum, 648 millimeters.
Steam pressure, about 0.7 kilo per square centimeter (10 pounds).

Experiment III.—An attempt was made to shorten the time of drying by increasing the steam pressure and having the machine hot before putting in the nuts, the initial temperature being 75°.

Actual time of drying, 5½ hours.
Maximum temperature, 85° C.
Vacuum, 640 millimeters.
Steam pressure, about 4.2 kilos per square centimeter (60 pounds).

Experiment IV.—The meat from four nuts was ground and spread in a layer of a depth of about 3 centimeters in shallow glass dishes. The initial temperature was 60°.

Actual time of drying, 9 hours.
Maximum temperature, 75° C.
Vacuum, 660 millimeters.
Steam pressure, 0.7 kilo per square centimeter (about 10 pounds).

Experiment V.—Four coconuts were split in halves and put into the machine, the latter being left partly open and with no vacuum.

Actual time of drying, 11 hours.
Highest temperature, 86° C.
Steam pressure, 0.7 kilo per square centimeter (about 10 pounds).

Therefore, under the best conditions obtainable (temperature 85° and vacuum 635 to 660 millimeters), the minimum time required for vacuum drying was five and one-half hours.[11]

If we are to form our judgment from the great efficiency of the vacuum evaporators used for sugar solutions and for many other liquids, it might be supposed that this process would be equally advantageous for coconuts. However, the two conditions are altogether different. In the case of solutions we have a thin layer of liquid in direct contact with a heated surface, the evaporation taking place so rapidly that the space above the liquid is constantly saturated with moisture; the main object of these machines is to remove and condense the surplus water vapor as rapidly as possible and by so doing to allow the evaporation to proceed at a comparatively low temperature. The water in coconut meat, on the other hand, which at the most is not greater than 50 per cent of the total weight of material, under the best conditions, diffuses very slowly through the cells of the copra to the surface, the removal of moisture-laden air therefore becoming a matter of secondary importance. The principal consideration is the constant application of as much heat to the entire surface of the material as the latter can endure without becoming burnt. That this condition is not fulfilled in the best manner by a vacuum drier is chiefly due to the poor conductivity of the rarified air which it is necessary to heat. Although the temperature of the steam plates in the drying oven is from 100° to 110°, that of the partial vacuum immediately above and surrounding the copra, even after several hours, rarely rises above 75°. To this local superheating for a long period of time at the point of contact with the plates, probably is due the brown color and slightly burnt taste which vacuum-dried copra almost invariably possesses.

For the sake of comparison I append the following table showing the

[11] This does not include the time necessary to produce steam and to heat up the drier. These items must be considered unless the apparatus is to run continuously.

approximate time required to dry copra under the most favorable conditions by each of the methods previously considered:

Method.	Time.
Sun	5 days.
Grill	10 to 12 hours.
Hot air (box)	3½ to 4 hours.
Hot air (rotary)	2 to 3 hours.
Vacuum	5½ to 6½ hours.

The quality of the copra produced by the hot-air box drier is very much superior to that yielded by any other method, since it is perfectly white and dry, retaining the pleasant odor and taste of fresh coconut meat. For oil-making purposes the rotary apparatus, because it lends itself to a continuous process and requires considerably less time, recommends itself especially, although its product does not present quite so pleasing an appearance. Either of these two methods, on account of their cheapness and simplicity, should be preferred to vacuum drying.

Centrifugating.—Another method of drying suggests itself, which should prove to be very efficient, although, owing to lack of facilities, I have not as yet been able to give it a practical test. This is to extract the meat from coconuts by means of a rotary burr and to run this product directly into a powerful centrifugal from which the greater part of the water would be thrown off at once. A comparatively short, supplementary drying by means of hot air would then suffice to prepare copra for expressing the oil. Another point in favor of this method is that the copra resulting therefrom, having lost most of its sugar and albuminoids together with its water in the process of centrifugation, would be able to withstand a higher temperature while drying (with a resulting economy of time) without showing the same tendency to turn brown. Once dry, it could be stored with less danger of deterioration through mold action than material prepared by ordinary methods. The objection may be raised that, during the centrifugation, a considerable amount of oil together with the water would be thrown off from the fresh meat, and that this would either entirely be lost or would necessitate much labor for its recovery. This, to a certain extent, is true, as the water in coconut meat exists in the form of a cream-like emulsion with oil, sugar, and albuminoids. A sample of this "coconut cream," prepared by expressing the fresh meat in a hand press, was, on analysis, found to have a specific gravity of 1.012 at 30° C. and to consist of—

	Per cent.
Water	56.3
Total solids	43.7
Ash	1.2
Fat	33.4
Proteid (N x 6.25)[12]	4.1
Total sugar as invert sugar	5.0

[12] Determination made by Mr. Richmond, of this laboratory.

The above results show that it approximates in nutritive properties the composition of a rich, natural cream; it is very pleasant and sweet to the taste, possesses an agreeable odor, and, when sterilized and properly sealed, will remain indefinitely in a fresh condition. Such a product could be used as as substitute for all of the purposes to which the so-called "evaporated creams," now on the market, are put, and it might prove to be one of the most valuable by-products of the coconut-oil industry.

THE ACTION OF ORGANISMS ON COCONUT OIL UNDER VARYING CONDITIONS.

Although, as has been shown above, the character of a coconut oil in regard to free acid, odor, and taste is determined chiefly by the quality of the copra used for its production, there is also in most commercial oils a slow but steady deterioration, amounting in the worst cases to a rise of about 0.5 per cent per month (cf. sample No. 31, p. 120), while with pure, filtered oils this reaches only a few hundredths per cent in the same time. It has been remarked above that samples of oil which contain suspended impurities and water, as a rule, increase in their content of free acid somewhat more rapidly than do similar ones which have been clarified; a result to be expected, if, as is the case with copra, decomposition is due to micro-organisms, since it has been proven that bacteria and molds do not live for any length of time in pure oil.[13]

The influence of impurities on the keeping qualities of oils was noticed as early as 1855 by Pelouze,[14] who observed that various oleaginous seeds, when crushed and extracted at once, yielded almost neutral oils, whereas if, after being crushed, they were allowed to stand for some time before extraction, the oil then produced contained a large amount of free acid. He considered this action to be due to a "ferment" similar to that producing alcohol from sugar.

Pastrovich and Ulzer,[15] using a mixture of oleomargarine with 0.5 per cent casein and 1 per cent water, observed an increase of acidity from 0.888 to 1.259 per cent in one week, and in fourteen weeks 0.888 to 10.270 per cent. They make no attempt to explain this effect, evidently attributing the saponification to some change brought about directly by the presence of albuminoids, although it is very probable that it was produced by bacteria or molds.

The following experiments were undertaken with a view of accentuating this difference in keeping qualities between pure and impure oils by exposing them directly to the action of micro-organisms under similar conditions.

About 20 cubic centimeters each of samples Nos. 6, 8, and 11 were poured into small beakers and placed in a covered specimen jar containing

[13] E. Ritsert: Untersuchungen über das Ranzigwerden der Fette. *Chem. Centrb.* (1890), 507, 575, 813.

[14] M. J. Pelouze: Memoir, sur la saponification des huiles sous l'influence des matieres qui les accompagnent dans les grains. *Compt. Rend.* (1855), **40**; 605.

[15] Pastrovich u. Ulzer: Ueber den Einfluss der Gegenwart verschiedener Eiweisskörper auf Fette. *Ber. d. chem. Gesell.* (1903), **36**, 209.

a little moldy copra together with a beaker of water to insure an abundance of moisture. They were then allowed to stand at room temperature (25° to 30° C.) for two months. Their change in acid value is shown as follows:

Number of oil.	After standing one month.		After standing two months.	
	F. F. A. in oil exposed to mold.	F. F. A. in oil in original bottles.	F. F. A. in oil exposed to mold.	F. F. A. in oil in original bottles.
6	0.22	0.16	0.26	0.17
8	0.28	0.14	0.26	0.17
11	0.22	0.18	0.88	0.22

A slight increase of acidity was evident in each of these samples, but No. 11, an unfiltered, very turbid oil, was decomposed much more rapidly than either of the others. It also was characteristically rancid in odor and taste and contained a visible mold growth.

Since a marked difference was shown in the behavior of these oils, it was decided to observe the effect of the addition of small quantities of nutrient matter and of water on the rise in the free-acid contents of a pure oil. The nutrient material which was used was prepared from "latic," a coagulated residue produced in the native process of making oil, by boiling down an emulsion of fresh coconut meat. This residue, when dried and extracted with chloroform, yields a light-brown powder, partially soluble in water and of a sweetish, not unpleasant, taste. It consists chiefly of albumin and sugar. The following samples were prepared, using pure, fresh oil as a base:

A. 25 cubic centimeters oil + 0.25 grams "latic."
B. 25 cubic centimeters oil + 0.25 grams "latic" + 0.25 cubic centimeters water.
C. Control of pure oil.

Each of these samples was placed in a 50-cubic centimeter glass bottle inoculated with one drop of the moldy oil (No. 11) used in the previous experiments, and allowed to stand for one week at room temperature, and, after determining the increase in acidity, for one week in the incubator at 35°. Finally, the acidity was tested after the oil was allowed to stand for two months longer at room temperature.

Sample.	Initial acidity.	One week at room temperature.	One week at 35° C.	Two months at room temperature.
A	0.10	0.11	0.13	0.27
B	0.10	0.25	0.69	3.8
C	0.10	0.10	0.11	0.13

The following table gives the results of a similar experiment in which 10 cubic centimeters of the oil to be tested were inoculated with a drop of moldy oil, then poured out in a Petrie dish, which was placed in a closed specimen jar over a little moist, moldy copra, and allowed to stand in the incubator at 33° to 35° C. for one week:

Nature of oil.	Initial acidity.	One week at 33°–35° C.
No. 29	0.10	0.50
No. 30	0.10	0.21
No. 30 + 1 per cent "latic" + 1 per cent water	0.10	8.63

The last sample, which showed the greatest increase in acidity I have yet been able to produce in a short time, was almost completely covered by a greenish-yellow mold, similar to that noticed on copra, and it had the characteristic odor of a rancid oil.

In the light of these latter experiments there can be no doubt of the fact that coconut oil, provided it contains sufficient moisture and nutrient matter, is attacked by micro-organisms, principally molds, with an accompanying production of free acid and of a disagreeable taste and odor. This is the principal cause of "rancidity" in coconut oil, if by "rancidity" we mean a high acid content and a bad taste and odor. Whether this fat splitting is directly due to a life process of the molds or to an enzyme secreted by them is a problem which has not yet been solved. However, it seems highly probable that these molds produce a slowly acting enzyme, soluble in oil, which continues its hydrolytic action even after the organisms themselves are dead. This would account for the steady increase in free acid of some commercial oils which are perfectly clear and free from impurities and which have been proven to contain no living bacteria or molds. Experiments are now being carried on to clear up this point.

OTHER FACTORS INFLUENCING THE ACIDITY OF OIL.

Effect of sunlight.—Twenty-five cubic centimeters each of samples "A" and "11" (see table, p. 119) were placed in 50-cubic centimeter glass-stoppered bottles and allowed to stand in the sun for one month. At the end of this time "A" contained 0.22 per cent and "11" 0.24 per cent of free acid, while at the same time the original samples "A" and "11" showed 0.06 and 0.18 per cent, respectively. No marked change in taste or odor could be detected. The acid content of the pure sample "A" appears to have increased considerably more than that of "11," due probably to its contamination with a few drops of water during a heavy rain. However, the total amount of acid developed was so small that the experiment was not repeated.

Effective of heat and moisture.—Several samples of oil were heated at 100° for periods up to twenty-four hours with no change in acid

value. However, on heating in a sealed tube, considerable alteration took place, as is shown in the following: Samples from No. "B" and from that portion of No. 11 which had been exposed to sunlight were heated in sealed tubes at about 160° for ten hours. "B" changed in percentage of free acid from 0.06 to 0.90, but was unchanged in color, odor, or taste. "11" rose from 0.24 per cent to 2.05 per cent under the same conditions, and possessed a very disagreeable odor and a nauseating taste. It also showed a considerable increase in color, probably due to decomposed albumin, etc. The change taking place in the sample was demonstrated to be a simple hydrolysis by heat and moisture. In the next experiment, both "B" and "11" (original samples) were dried very carefully by passing through them a current of dry air for seven hours at 100°; 10 cubic centimeters each of the dry oils were then sealed in glass tubes and heated at about 160° for ten hours. For comparison, two more tubes of oil were subjected to the same conditions, one containing 10 cubic centimeters of sample 11 (undried), the other 10 cubic centimeters of the dried oil "B" plus 3 drops of water.

Oil.	Before heating.	After heating.
"B" dry	0.07	0.09
"B" + aq	0.07	14.8
11 dry	0.18	0.19
11 original	0.18	0.46

In neither of the samples marked "B" was a bad odor or any color produced, though the hydrolyzed sample was slightly unpleasant to the taste, owing to the large amount of free acid present. However, both the similarly treated tubes containing oil No. 11, in spite of their low acid value, were decidedly disagreeable to the taste and smell and presented a decomposed appearance.

FACTORS WHICH CAUSE RANCIDITY IN OIL.

The average person, if asked to judge of the quality of these four heated oils by the sense of taste and smell alone, would almost invariably say that both samples of oil No. 11 were "rancid" and that the other two were fairly pure, whereas, judging from the amount of free acid present, one might consider "B + aq." as the only sample containing a marked amount of rancidity. From the above it is evident that rancidity and free acid are not by any means synonymous and that the cause of the former must be sought elsewhere than in the percentage of the latter.

Lewkowitsch applies the term "rancidity" only to those fats which contain an excess of free fatty acids due to the action of air.

Alder Wright[16] gives a résumé of the work done on this subject and concludes

[16] Alder Wright: *Fixed Oils, Fats, Butters, and Waxes*, second edition (1903), 168.

that rancidity is the alteration which oils and fats undergo when not protected from the influence of *air and light*. "Such oils," he says, "acquire a sharp, disagreeable taste and odor, their proportion of free acids gradually increases, and they undergo various other chemical alterations."

Schmid[17] differentiates between "sour fats," "rancid fats," and "sour and rancid fats." "A fat is sour," he says, "when its content of free fatty acids is abnormally high but the free glycerine is unchanged. A fat is rancid when the proportion of free fatty acids is not high but the free glycerine has been oxidized partially or completely to aldehydes and ketones. A fat is rancid and sour when it contains a large amount of free acid together with oxidation products of glycerine." As a test for rancidity he proposes a 1 per cent solution of m. phenylendiamin.

Scala[18] found œnanthylic aldehyde among other substances present in rancid olive oil, and assumes that this body gives the characteristic odor and taste termed "rancidity."

Bianchi[19] proposes, as a test for rancidity, to shake up a little of the oil in question with fuchsin-sulphurous acid, a violet-red color indicating rancidity.

Brown[20] uses this same test in the study of butter fat, and presumes that the rancid odor is due to acrolein. Various other tests for rancidity have been proposed, all depending on the presence of aldehydes.

The most satisfactory, in my experience, is that with fuchsin-sulphurous acid, shaking up about equal parts of oil and reagent. Nearly all the samples of coconut oil prepared in this laboratory, after standing for several months, responded to this test, but they gave, not a violet red, as has been stated to be the case with other rancid oils, but a more or less blue coloration with only a slight tinge of red. The above review of the literature will demonstrate that the causes of rancidity are by no means clear. Certainly œnanthylic aldehyde has not an intensely disagreeable odor; acrolein has, but then it gives a red and not a blue color with fuchsin-sulphurous acid.

ACTIVE OXYGEN IN COCONUT OIL WHICH HAS BEEN STANDING.

Another peculiarity of pure coconut oil is that, after it has been standing exposed to light and air for a few months, it almost invariably contains active oxygen. Five cubic centimeters of sample "A," shaken in an Ehrlenmeyer flask with a mixture of 50 cubic centimeters of water, 5 cubic centimeters glacial acetic acid, and 1 gram potassium iodide and allowed to stand for one hour, produced a deep-yellow coloration in the water solution, requiring 0.25 cubic centimeter $\frac{N}{10}$ sodium thiosulfate for decolorization. A blank test, with freshly prepared oil, remained perfectly colorless during the same time. A simple way of performing this test is to saturate a strip of starch iodide paper with the oil in question,

[17] A. Schmid: Zur Prüfung der Fette auf Ranzidität. *Z. Anal. Ch.* (1901), **37**, 301.

[18] Alberto Scala: *Staz. sper. Agrar. ital*, **30**, 613, *Centrbl.* (1898), 439.

[19] *Centrbl.* (1898), II, 948.

[20] Brown: The Chemistry of Butter Fat. *Jour. Amer. Chem. Soc.* (1899), **21**, 975.

place it on a glass plate, and carefully add one drop of 10 per cent acetic acid. When old oil is used, a blue ring appears around the drop of acid in from one to five hours, whereas a freshly prepared sample remains uncolored for twenty-four hours or more, provided the test paper is kept under a bell jar to exclude laboratory fumes. This reaction is at least as delicate as any of the tests for rancidity based on the presence of aldehyde. Whether it is given by other rancid oils can not be stated at present, for I have as yet failed to find in the chemical literature any mention of such oxidizing substances in oil. The following is a table showing the reaction of our samples of coconut oil with fuchsin-sulphurous acid and with starch iodide, together with their age and acidity at the time of testing. For the previous history of these oils, see page

No.	Approximate age, in months, at time of testing.	Approximate percentage F. F. A.	Color with fuchsin-sulphurous acid.	Test for peroxides.
A	15	0.1	Strong blue	+*
B	5	0.1	do	+*
1	5½	1.5	do	+
2	5½	1.5	do	+
3	5½	2.1	do	+
4	5½	5.9	do	+
5	5½	7.6	Blue-red	—
6	5½	0.2	Strong blue	Trace.
7	5½	0.2	do	+
8	5½	0.2	do	+
9	5½	0.2	do	+
10	5½	0.2	do	+
11	5	0.3	do	+
12	5	0.1	do	+
13	5	0.1	do	+
14	5	4.0	No color	Trace.
15	5	0.9	Strong blue	+
16	5	2.0	do	+
17	4	0.1	do	+
18	4	0.2	do	+
19	4	1.2	do	+
20	4	0.7	No color	+
21	4	0.7	do	+
22	3½	23.3	Red	—
23	3½	1.6	Light blue	Trace.
24	3½	1.5	Strong blue	+
25	3½	3.4	Moderate blue	+
26	3½	2.6	Strong blue	+
27	3½	2.4	Light blue	Trace.
28	3½	3.5	Very strong blue	+
29	2	0.1	No color	+
30	2	0.4	Light blue	+
31	3½	3.5	Trace	+
32	3½	7.8	Moderate blue	—
33	7	7.0	No color	—
34	5½	10.2	Strong blue	+
35	½	5.0	No color	+

*Very strong.

In examining this table it will be noticed that in almost every case a distinct blue color with Schiff's reagent is accompanied by a positive test for peroxide, and, in the two cases where a red coloration predominates, the peroxide test is negative. The strongest tests for both peroxide and aldehyde were given by samples A and B—two very pure oils—while, contrary to our expectations, the commercial oils as a rule either failed to respond entirely or gave very weak tests, though they were infinitely worse than the pure samples in every other particular. The only change noticeable in samples A and B, on standing, was the development of a peculiar, pungent, "strong" odor and a slight burning "after taste;" otherwise they were practically the same as when freshly prepared. Just what this "strong" odor in pure oils is caused by can not at present be stated, although the subject is now being investigated. It can hardly be caused by either œnanthol or acroleïn, as both these substances, when mixed with oil, give a red and not a blue color with Schiff's reagent. Glycerine aldehyde[21] under certain conditions produces a blue-red coloration with this test, but it has no odor. However, the process by which this change is produced is undoubtedly due to direct oxidation by light and air, since bacterial or mold action may be excluded in the case of a pure oil. That it is largely a surface action is indicated by the facts that (1) samples A and B, which were kept in large bottles, about half full, have deteriorated in five months to a much greater extent than have other samples of pure oil which were kept in small, nearly full bottles for over a year; (2) both the aldehyde and peroxide tests were given by samples of fresh oil which were exposed to the air on strips of filter paper for one or two weeks;[22] (3) the same effect can be produced by treating fresh oil with platinum black for a few hours, or by heating it, exposed in a thin layer, to 100° for ten or twelve hours.

A possible explanation of this production of rancidity in pure oils is that a small percentage of fatty acid is oxidized to an oxyacid, which in turn forms a lactone, and (assuming the formation of hydrogen peroxide) the latter would give rise to a peracid, which, in turn, would oxidize the free glycerine to an aldehyde. The absence of peroxide and, as a rule, of aldehyde in commercial coconut oils, or in those purposely subjected to the action of micro-organisms, may be due to the presence of sugars and other reducing substances commonly present in the impure oils, or to the fact that the glycerine set free by mold action is completely oxidized to carbon dioxide and water. The nonexistence of free glycerine in highly rancid fats has been noted by Sparth[23] and other observers. This question of the products arising from the oxidation of pure coconut oil by air is now being taken up more thoroughly, and the results will be published in a later paper. However, from a commercial point of view, it is of

[21] E. Fischer u. Tafel. *Ber. d. Chem. Gesell.* (1887), **20**, 3384.
[22] Freer & Novy: *Amer. Chem. J.* (1902), **27**, 161.
[23] *Zt. Anal. Ch.* (1896), **35**, 471.

comparatively little importance, when one considers the marked deterioration produced by micro-organisms acting on copra and impure oil. If stored in nearly air-tight containers very little if any oxidation should take place even on long standing or on transportation. The main points to remember are that the copra from which oil is made should be fresh and be prepared under as good conditions of drying as possible and the oil should be thoroughly dried and filtered until absolutely clear. If properly prepared, it should then be capable of shipment without noticeable deterioration. It is obvious that the best results will be obtained by expressing the oil in the country in which copra is dried, and by using the best machinery for preparing the latter.

SUMMARY.

Soil.—In attempting by means of soil analyses to explain why coconut trees growing near the seashore are more prolific than those planted farther inland, it was observed that—

(1) Chemically, there is very little difference in soils from the two localities, those from inland regions being, if anything, a little more fertile.

(2) The salt water from the sea has no influence on trees in its vicinity, as only amounts of chlorine so small as to be negligible were found to be present even at the bases of coconut trees which were actually growing on the beach.

(3) The greater porosity of soils near the sea, coupled with the fact that they are, as a rule, practically saturated with water at a distance of only a few feet beneath the surface of the ground, is the principal reason why they are more suitable for trees like the coconut, which require an enormous quantity of water for their growth.

(4) Although good coconut soils are apparently almost devoid of fertility, yet, taking into account the character of coconut roots and the large area from which each tree draws nourishment, it can be demonstrated that there exists an ample supply of nutriment for their growth.

The nut; age in reference to quality.—(1) The variations among individual nuts is sufficiently great to render exact conclusions from analytical data difficult, but, taking the average of a number of determinations, there appears to be a slight increase in the proportion of meat, copra, and oil in nuts which have been stored up to a maximum time of three months after cutting. Beyond this period there is a decided decrease in these constituents. Nuts taken from the same tree show somewhat less individual variation.

(2) Four series of ten nuts each, of varying degrees of ripeness, showed a marked difference in the amount of copra and of oil to be obtainable from them, the percentage of the oil in a green nut being only about one-half of that which it is when the nut is fully ripe. This ripening process continues to some extent, on storage, after cutting.

(3) Analyses of coconuts from the same locality, but having husks of different color, prove that the color of a nut has very little if any influence on its composition.

(4) The difference between trees near the seashore and those farther inland is solely in the quantity, not in the quality, of nuts which they produce, coconuts from inland regions averaging fully as well as those from the beach. This fact is shown both by analyses and by practical tests on a large scale.

(5) Analyses were made of twenty ripe coconuts from Davao, Mindanao, and they were found to have very much the same proportion of the various constituents and to give the same total yield of oil as the average lot of ripe nuts from San Ramon.

(6) Coconut oil is generally stated to have a great tendency to become rancid, but all the experiments made in this laboratory show that, when once prepared in a pure state, its keeping qualities are equal if not superior to those of most other vegetable fats and oils. This popular fallacy in regard to coconut oil probably arose from the inability or disinclination on the part of most observers to procure pure samples, as the commercial product unquestionably has a high acid value and a bad odor, and deteriorates with fair rapidity, this change being greater as a rule the greater the initial acidity of the oil.

(7) Most of the free acid and the accompanying bad odor and taste is produced in the copra itself before the oil has been expressed. The oil from a sample of copra which had been cut into fine pieces and exposed to moist air for one month increased in acidity from 1.5 to 23.3 per cent.

(8) The hydrolysis and subsequent destruction of fat in copra is brought about by molds (the greater part of which are *Aspergilli*), acting either alone or in symbiosis with certain bacteria, the condition most favorable to this growth being a moderately high, constant temperature and a water content of from about 9 to 17 per cent. No organisms were found growing on a sample containing 4.76 per cent of moisture and no change in acidity took place. Samples containing from 23 to 50 per cent of water were infested by several species of bacteria which subsisted on the nonfatty portion of the copra but produced very little free acid from the oil. No molds were found in these samples.

(9) Ordinarily, commercial copra contains from 9 to 12 per cent of moisture, a very favorable condition for mold growth. The remedy for this rapid deterioration is simply to dry it so that it contains not more than 5 per cent of moisture, and express the oil as soon as possible, avoiding long storage in a warm, moist atmosphere.

Drying.—By comparing the various methods of copra drying, a hot-air apparatus, either rotary or stationary, was found to be the most efficient. It is suggested that a combination of centrifugal with hot-air drying might prove of considerable value, provided a market could be

obtained for the by-product, "coconut cream." Vacuum drying is not of great value in the desiccation of coconuts for oil-making purposes.

(10) Although a pure coconut oil is not a suitable medium for a growth of micro-organisms, one containing a sufficient amount of nutrient matter and moisture may, under certain conditions, develop a growth of mold which rapidly attacks the oil itself. A sample of pure oil to which had been added 1 per cent of "latic" and 1 per cent of water increased in acidity from 0.10 per cent to 8.63 per cent on standing exposed to mold action in an incubator for one week.

The very slight increase in acidity which a pure oil suffers on long standing is probably due to simple hydrolysis by heat and moisture.

(11) Besides the production of free acid by molds and the decomposition of albumen by bacteria in moist copra and in impure oils, one other factor enters into the deterioration of coconut oil. Many samples on long standing develop a slight but noticeably acrid taste and odor, without any marked increase in acidity. Such oils invariably give a blue coloration with Schiff's aldehyde reagent, reduce silver nitrate in Becchi's test for cotton-seed oil, and possess the power of liberating iodine from potassium iodide.[24] This process is shown to be a direct oxidation by the air and to depend largely upon the amount of surface exposed. Other conditions favoring it are freedom from moisture and impurities, as is shown by the fact that impure commercial oils, or those which have been acted upon by mold, do not, as a rule, respond to tests for peroxide and aldehyde, while the most marked development of these bodies is noticed in the purest oils.

(12) The action of light and air on coconut oil is of relatively little importance in comparison with the great changes produced by mold growth, and it can be prevented in a large degree by keeping oil receptacles as nearly full as possible, so as to reduce the amount of surface exposed.

[24] Since writing the above I find that L. Legler [*Pharm. Centr.-H.* (1904), 45, 839] has noticed the same phenomenon in oxidized lard. As a test for active oxygen he proposes to shake the sample with a solution of neutral lead acetate and a few drops of ammonia. A yellow coloration, due to the formation of hydrated lead peroxide, indicates the presence of oxygen. I have applied this test to old coconut oils and find that, with highly oxidized samples, it gives a strong coloration, but it is not as delicate as the simple reaction with potassium iodide. To the presence of active oxygen Legler attributes the reduction of silver nitrate in Becchi's test given by samples of oxidized lard entirely free from cotton-seed oil. I have observed the same fact when applying Becchi's test to pure, but oxidized, samples of coconut oil, but considered it more logical to attribute the reduction to aldehyde-like bodies present in the oil, rather than to the active oxygen.

THE PRINCIPAL INSECTS INJURIOUS TO THE COCONUT PALM (PART I).

By CHARLES S. BANKS.

(*From the Entomological Section, Biological Laboratory, Bureau of Science.*)

Among commercially valuable trees, few are attacked by as small a number of insect pests as the coconut (*Cocos nucifera* L.); but, on the other hand, the destructive action of this limited number is very great.

The trunk of the coconut does not have its important conducting tissues in or immediately under the bark, as is true of the cacao, the coffee, or the mango. For this reason, even though the tree were completely girdled, it would not be destroyed, as would be the case with the plants above mentioned. On the other hand, insects attacking the growing point would soon kill this part, after which the remainder would speedily die, and, in fact, this result is the one which almost always is encountered. Certain insects enter the crown and destroy it; shortly afterwards, the leaves turn yellow, the fruits, if any are present, drop off, and the tree eventually dies. It is therefore clear that any method which prevents attacks of this kind will preserve the life of the tree.

This paper will treat of some of the most important of the insects destructive to the coconut which have been identified, while those the habits and life histories of which are known but the determination of which has not yet been made will be a subject for further study. The observations upon the habits and life histories have been made both in the laboratory and in the field. I wish to take this opportunity of thanking Mr. W. Schultze for his hearty coöperation in this work and for the illustrations which he has furnished.

THE RHINOCEROS BEETLE.

Oryctes Rhinoceros L. (Tagalog, *Uang*).

This insect belongs to the family Dynastidæ, or that of the giant beetles, a group in which, from the standpoint of body weight, the largest of the beetle tribe are found. This beetle is very common throughout the Philippine Archipelago and in other countries of the East (Ceylon, Java, India, etc.) wherever the coconut tree is encountered. All of the

species of the genus *Oryctes* apparently have the same predilection for the coconut and for similar palms. The presence of the rhinoceros beetle is indicated by the large, irregular holes in the trunks of the trees or at the bases of the largest petioles of the leaves of the coconut. These are made by the adult beetles and serve as a means of entrance for other insect pests, such as the Asiatic palm weevil, and also for the admission of moisture, which eventually causes the trunk to rot. The beetles' attacks are confined to the soft tissues near the top of the tree, and holes seen in the trunk below this point date from the time when the growing apex was here located.

No *Oryctes* has ever been found gnawing the hard, old wood of the trunk of the coconut; occasionally adult beetles are found in these old holes, which, however, are used only as a hiding place during the day. In some of them old cocoons which were constructed when the hole was at the crown of the tree are occasionally found, consisting of masses or bundles of fiber. These have been preserved *in situ*, because of the small size of the opening of the burrow. As time goes on, the old holes become enlarged through various agencies, particularly through erosion and decay caused by the entrance of water, so that these bundles of fiber finally become exposed.

Life history and habits.—Like all other members of the family Dynastidæ, *Oryctes* is a vegetable feeder. While it frequently occurs in heaps of decaying vegetation, the larvæ appear to have the greatest liking for the soft, growing point of the coconut, which is the location from which new leaves, the flowers, and, subsequently, the fruit, obtain their nourishment. Therefore, any injury to this part of the tree immediately results in debilitating the whole plant and eventually in its death. The mode of attack most generally encountered is that in which the female has entered between the long stems or petioles of the outside leaves and those immediately subjacent, and then has eaten a hole into the outer side of the inner petioles, which are protected from the light by the external leaf stems. As this beetle shuns the light, its attacks always begin during the night, and by the following morning it will frequently have entered so far into the burrow as to be protected from the light. It then continues its feeding until a gallery of considerable size has been excavated. The habit of burrowing would seem to be not solely for the purpose of laying eggs but also in order to obtain nourishment, as nearly all of the members of this family feed in the adult stage.

The egg.—I know of no record regarding the actual deposition of the egg, nor have I found it in any of the burrows from which the adult has been taken, but, by dissection of the females, the eggs have been obtained in considerable numbers. Just before being laid they are of a dark cream color and present a perfectly smooth texture. The microscope reveals a very delicate reticulation or punctuation of the surface. They are 3.5 millimeters long and 2 millimeters in diameter, being of a perfect

broad, ellipsoidal form. The structure of the ovipositor of the beetle leads one to infer that the egg is simply dropped by the female at any spot in the gallery, where it adheres to the side of the latter until the larvæ are hatched. In none of the females which I dissected were there more than seven or eight eggs of such a size as to indicate that they were about to be laid, so that the insect probably deposits not more than two dozen during its life. (Pl. I, fig. 1.)

The larva.—Length over dorsum when full grown, 112 millimeters; circumference, 18 to 20 millimeters; head, 12 millimeters long and 11 millimeters wide; feet, 9 millimeters in length. It is a very soft, fleshy grub, the skin of which is transversely doubled in numerous folds, so that it is very difficult to differentiate the body segments. The skin is of a dirty, light ocher, the surface being smooth in spots and in other parts covered with patches of very minute tubercles or spines, which give it the appearance of shagreen. The body is covered with numerous, very fine, golden or dull-whitish, curved hairs, which stand out nearly at right angles to the surface. The head is of a much darker color than the body and horny or chitinous in structure; it is hemispherical and so attached to the body that the mouth projects forward. At each side of the head, projecting downward and forward, is a slender, four-jointed antenna. The dark-brown, slender, toothed mandibles, half the length of the entire head, are so placed as to enable the insect to gnaw its way through the plant substance with great facility. The maxillæ are rather conspicuous and are situated next to the mandibles on the under side of the head and on each side of the very inconspicuous lower lip or labium. The maxillary palpi are 4-jointed; those of the labrum 2-jointed. The labrum is transversely elliptical, the sides slightly covering the inner margins of the mandibles. The color of the labrum and that of the clypeus (the trapezoidal portion above the upper lip) is the same as that of the rest of the head. The mouth parts, with the exception of the tips of the mandibles, are covered with golden bristle-like hairs, which serve as tactile sense organs.

This grub has no eyes. The top and front of the head, therefore, present an unbroken surface, which is somewhat shiny and covered with punctures, which are almost confluent; it is nearly destitute of hairs or bristles. (Pl. I, fig. 2.)

Each of the first 3 segments of the body posterior to the head bears a pair of legs. The first leg joint projects downward, while the succeeding ones are inclined outward and forward; the feet are armed with a single blunt claw and densely covered with light-brown bristle-like hairs, more thickly placed at the tips.

The body is curved, so that the length of the ventrum is much less than that of the dorsum. It is folded or transversely corrugated so as to render it difficult to distinguish the 13 segments of which it is composed, this being the easier toward the anal extremity, where the folds are fewer.

Beginning with the first thoracic segment and excepting the second and third, each remaining one to the eleventh bears a pair of dark-brown subcircular spiracles or openings to the tracheal or respiratory system. These spiracles are chitinous in structure and are composed of an outer broken ring of radiating lines and an inner nearly circular portion, which is the true opening. They may be opened or closed at will. Their large size and great prominence is doubtless owing to the fact that the insect lives embedded in a mass of material in which the supply of oxygen is limited.

The last three segments of the grub's body are nearly smooth and only sparsely covered with hair, and in a living specimen the hinder end is semitransparent

and contains a dark mass of material, consisting of the partly digested cellulose fibers of the plant upon which the insect has fed. The anal opening occurs as a transverse slit at the extremity of the body. (Pl. IV, fig. 1.)

Differences of opinion appear to have existed with reference to the destructiveness of the grub of *Oryctes*. Blandford says:

They are harmless, and live in heaps of rotting vegetable matter or the manure-like inside of decayed palm trees.

Both Mr. Schultze and I have discovered them in large numbers in coconut trees in which the "manure-like" material inside the trunks gave every evidence of having been made by the grubs themselves. One tree, felled in the town of Magdalena, Laguna Province, while still alive and to casual observation fairly healthy, was found to have an inverted cone eaten out at the crown, as shown by Plate III, fig. 1. This contained seven of the grubs of *Oryctes rhinoceros* L. buried in the frass. There was a tunnel, 3 centimeters in diameter, extending down from the apex of the cone for a distance of 90 centimeters through the heart of the tree, and at the bottom of this tunnel was a full-grown grub, which to all appearances had eaten its way to this point. Mr. Schultze observed in Pagsanjan a tree (Pl. II) 5 meters high, the whole of the interior of which had been eaten out from its top to within a half meter of the ground, leaving a shell with a wall from 15 to 20 centimeters in thickness. Within this, at the lower part, was a mixture of water and decayed matter 50 centimeters deep, indicating that the work of *Oryctes* and the Asiatic palm weevil, together with infiltration from the top, had been continuing for a considerable period of time. Within this rotting mass and at intervals up to the crown of the tree were found the fiber cocoons of *O. rhinoceros* L., while from 75 to 100 larvæ of all sizes, from 5 millimeters to the full-grown grub, were removed. The small number of weevils, the large number of *Oryctes* larvæ and pupæ, and the general appearance of the interior of the tree furnished conclusive proofs that the work was that of the insect in question. Leaving these points aside and reasoning from the anatomy of the larva alone, it is evident that it could work in the wood of coconut with great ease, since it is in every way fitted for burrowing there. If it lives only in manure heaps or in decaying matter, it would appear that there would be no necessity for such well-developed and powerful mandibles, nor would the head have to be of such hard material. It is true that these grubs are always encountered in the presence of decayed matter, either in the tree or in manure and other vegetable heaps, but, when found in the tree, it is probable that the decaying material is a result and not the cause of their presence. It is also true that we never cut into a tree until it shows unmistakable signs of insect attack or disease, and therefore do not see the work of the beetle at its incipiency. I have seen larvæ one-quarter grown removed from the burrows made by the adults in small coconut trees, the leaves of which were pulled apart; and I have also observed the

removal of grubs with a piece of bent wire from young coconut trees, although at the time I did not examine the hole sufficiently to note the actual work of the larvæ.

However, it is not to be doubted that these same insects find a suitable place in heaps of decaying vegetable matter, as the grubs have been found in such locations in all stages of growth. In connection with this question, Father Stanton, formerly of the Manila Observatory,[1] makes the following observations:

> We have found several live pupæ in a partly decayed stump of *Pithecolobium saman* that had been lying on an old wood pile for months; at another time we discovered one in a neat oval earthen cell within a broken bottle lying in a heap of refuse near a stable; and on one occasion, in a single heap of earth and manure, within a space of 1 cubic yard, we gathered dozens of larvæ in all stages of development from specimens 1 centimeter in length to those of 12 centimeters just about to transform to pupæ together with half a dozen pupæ and as many perfect beetles with their elytra still rather soft, as though the insects had just emerged from the pupal envelope. In this latter case, at least, it appears quite evident that the whole cycle of the metamorphoses of the insect took place right in this small pile of manure or very near to it. For, as many of the larvæ were very young, they could not well have migrated from the interior of a coconut or buri palm, seeing that there was not a single one of these trees in the whole neighborhood. It is evident then that *O. rhinoceros* does sometimes pass its whole larval and pupal existence in the midst of decayed or decaying organic matter, and consequently that the eggs are deposited in such situations. Whether they are also laid in the holes made by the female in the living tree is still to be ascertained, though from the fact that the grubs are sometimes found feeding in the heart of the tree high up near the crown it seems quite probable.

He quotes Señor Vicente Reyes, of Santa Cruz, Laguna, who says:

> It is remarked that coconut trees with all the leaves fresh, with blossoms and fruit all in perfect condition, and without any apparent cause, fall to the ground as though a hurricane had cut them down. On being examined it is found that from the roots up to the distance of a meter above the surface they are completely hollowed out, the whole interior having been converted into a mass of sawdust, and ensconced therein are a number of these worms, which have entered from the roots and worked upward, little by little, eating away and living upon the substance of the trunk itself.

In every case where I have encountered *Oryctes* in trees, except in those which were completely hollow, the work evidently proceeded from above downward. Of course, in those which were hollow, the channels of the grubs were found along the inner surface of the shell of the tree, but the evidence thus exhibited was not conclusive as to whether the larvæ had worked from above downward or the reverse.

Father Stanton notes the finding of the larvæ, pupæ, and adults of *Oryctes* in manure and other decaying organic matter, but he also says that he has not ascertained whether the eggs are laid in the holes made by

[1] *Phil. Weather Bur., Bull.*, August (1903), 225.

the adult beetles in the trees, adding that the fact that the grubs are sometimes found feeding in the heart of the tree high up near the crown makes it seem quite probable that the eggs are laid there also. Every evidence in my experience points to the great likelihood that they are laid in these holes.

When a tree which is so badly infested as to give external signs of debility is cut down, one usually finds larvæ of all stages as well as pupæ. The question of the length of the life period of this insect is a difficult one to determine, but from examinations, such as it has been possible to make during the time the insects of coconut have been under special observation, I am led to believe that it varies from eighteen months to two years, according to the food conditions. These conditions are determined largely by the size of the plant and the proportional number of insects in it.

Pupa.—The pupa of a female measures about 50 millimeters in length and 25 millimeters in width. The distance over the back from the tip of the head to the hinder part of the body, which in the pupa is curved forward, is 65 millimeters. It is of a light ocher yellow, in certain lights presenting a bright satin sheen and in others a velvety appearance. The head, thorax, abdominal segments above and below, and the wings and legs are all plainly visible, the anterior apex of the pupa, at a point corresponding with the top of the head in the adult insect, shows a small sharp knob or tubercle, which represents the horn of the full-grown beetle. A very fine golden pubescence, covering certain areas of the pupal body, causes its velvety appearance. The spiracles are placed similarly to those of the larva, but are almost hidden by the folds or wrinkles of the abdominal segments. On each side of the middle line of the back of the abdomen, transverse slits, very much like spiracles in appearance and undoubtedly secondary breathing orifices, are seen. These occur between each two abdominal segments, beginning with the first and continuing to the seventh, inclusive. Between the seventh and eighth there is indication of their existence in an atrophied state. (Pl. IV, fig. 2.)

Cocoon.—The cocoon is composed of fibers of the coconut, wound transversely and rather compactly woven or matted together. It sometimes measures 100 millimeters in length and 40 millimeters in diameter. When these insects live in rotting material or manure, the cocoon consists simply of an oval excavation, the interior being smoothed by the larva previous to its transformation. Unlike many pupæ of insects which feed in the interior of masses of material, this one has no organs by means of which it may cut or push its way out of the cocoon at the moment of transformation to the adult.

Adult.—The full-grown insect varies in length from 34 to 48 millimeters, according to the sex and the amount of nourishment taken in the larval stage, the average for the males being 44.2 millimeters and that for the females 37 millimeters. They are of a very dark-brown, somewhat lighter beneath, and have a very glossy or shiny appearance. The most striking feature is the horn on the fore part of the top of the head, this being much larger in the male than in the female. The head, thorax, and abdomen are easily distinguished. (Pl. IV, fig. 3.)

Male.—The head, with the exception of the horn, is irregular in form and subglobose; the front is strongly emarginate or sulcate. It is small in comparison with the thorax and so concealed posteriorly by the thorax, into which it fits very snugly, that it appears to be subtriangular from above. The eyes

are black and shiny and so situated at the sides of the head as to be half concealed by the anterior margin of the thorax. The anterior margin of the orbit is extended so as almost to cut the eye into upper and lower sections. The portion of the head at the base of the horn, which extends upward directly from the clypeus, is very densely pilose or setose, as is also the frontal sulcus. The occiput has an emargination at each side of the median line; into these fit the strong tendons of the powerful muscles which move the head upward. The antennæ project from the under, outer margin of the head. They are composed of 11 segments, the apical 3 of which are laminate; the first is swollen at the apex, is as long as the succeeding 7 together, and is very strongly pilose, the bristles being on its anterior, external surface (Pl. I, fig. 3). The ninth and eleventh segments are also pilose at their outer margins and tips; the tenth, lying concealed between them, is smooth and blade-like. The small, 4-jointed maxillary palpi lie just beneath the insertion of the antennæ at each side of the labium, which is subquadrate, with the anterior surface strongly swollen. Its lateral margins are strongly setose. The maxillæ are laminate and hidden between the labium and the peculiarly shaped mandibles. They are strongly setose on their exterior margins. The 3-jointed labial palpi lie beneath or anterior to the maxillary palpi and are attached to the apical part of the lateral margin of the labium.

The most peculiar feature of these insects, which has hitherto been unmentioned in the literature on the habits of the adult, is the special form and function of the mandibles. In the general description of the genus to which this insect belongs, the statement is made that "the mandibles are prominent and sometimes toothed externally." In the rhinoceros beetle the external tooth of the mandible is curved upward and forward and has the form of the cutting edge of a nonconcave gouge. These teeth, one on each side of the head, are by their construction and that of the surrounding parts well adapted for chiseling out the wood of the tree. (Pl. I, fig. 4.)

The shape and position of the external mandibular teeth, the form of the mentum, or chin, which is rounded and curved vertically, and which fits into a groove having a like form, in the anterior margin of the prothorax, together with the strong, well-attached muscles at the back of the head, prove conclusively that the insect, instead of gnawing its way into the tree, chisels into it by an up-and-down motion of the head, and it is my belief, for reasons to be given later, that no part of the wood is taken into the body.

The horn of the male is 10 millimeters in length and 4 millimeters in width at its base, tapering gradually to 1 millimeter at the tip, which in many specimens appears as if worn off and repolished. It is sparsely punctured, these punctures being fewer toward the tip.

Thorax.—The pronotum occupies about one-third of the length of the insect on the dorsum, is roughly subcircular in general outline, narrower anteriorly, having the margins somewhat reflexed. The anterior two-thirds shows a large, shallow depression, the surface of which is transversely or concentrically striopunctate, and at the posterior margin of which are two rather inconspicuous tubercles, almost coalescing. On each side and in the forward angle of the pronotum there is an irregular depression, posterior to which and extending narrowly around the posterior margin of the main depression, is another parenthesis-shaped one, broader anteriorly and having its surface roughly rugose. A line of submarginal punctures extends around the pronotum. (Pl. I, figs. 5 and 6.)

Elytra.—At the base, the wing covers are as wide as the thorax, the surface at the outer basal angles being quite smooth. On each elytron are four lines, one of which is subsutural, extending from base to apex. The external ones are

very indistinct toward the apex. The part between these lines is coarsely punctured, the punctures being regular on each side of the three external lines and on the exterior of the subsutural ones. The triangular scutellum, between the bases of the elytra, is smooth at its apex. There is a triangular rugose or coarsely irrorated area at its base.

The under surface of the thorax is of a chestnut brown; it is highly polished on the areas against which the legs move and strongly punctured on other exposed parts, the punctures having a sparse pile of light-brown bristles.

Legs.—The femora are uniform in size and smooth, each with 1 row of setose punctures nearer the posterior ventral margin. The tibiæ are nearly similar in shape and size, bearing externally 3 rather prominent teeth. The fore tibiæ, in addition, have an internal and external apical one. The mid and hind tibiæ have each 1 internal and 2 external apical teeth, armed with a row of smaller secondary ones, and all are coarsely punctured. All the tarsi are of about the same shape and size, except that the last joints of the anterior ones are slightly longer than the others, and the first of the mid and hind tarsi are subconical and slightly larger than the succeeding ones. All tarsal joints are setose or spinose at their apices.

Abdomen.—The dorsal abdominal segments are hidden by the elytra; the 6 visible ventral ones are smooth, except for very sparse punctures and a subapical row of setose punctures on each. There is a tuft of brown hairs at the anal slit. The hinder exposed part of the abdomen is rounded, smooth, shiny, and sparsely punctured. The elytra do not cover the last 2 dorsal segments.

The principal differences between the female and the male are that the former is much smaller and its horn may be a mere tubercle, or, at best, not more than one-fourth as long as that of the male. The depression on the pronotum extends back less than halfway; the posterior lateral rugose areas are somewhat broader. (Pl. I, figs. 5, 6). The last ventral abdominal segment of the female differs from that of the male, in that in the former it is rounded and covered with bristly hairs, while in the latter it is markedly emarginate. The ventrum of the abdomen of the female is rather densely covered in transverse rows with bristles, except along the apical margins of the segments. The posterior part of the pygidium is also densely hairy. Both sexes have on all the thoracic joints, as well as at the articulation of the head with the thorax, a fringe of bristles closely applied to the surface upon which the part is attached to prevent the entrance of foreign matter between the sutures.

Method of operation of the adult.—The method of attack of the adult insect was formerly believed to consist in its gnawing into the plant for the purpose of feeding upon the soft tissues inside, its eggs not being laid in the tree. This view is partly incorrect. Observation has shown that the males make burrows, as well as the females, and it is probable that they always accompany the latter at the time of egg-laying, retreating from the burrow they have made to allow the female access. Dissection demonstrates that the stomach of the insect contains no masticated fiber; on the contrary, it is filled with a dark, amber-colored liquid; nor are there any fiber cells found in the excreta. The proventriculus or gizzard of the insect is not provided with walls for grinding and the mandibles are constructed somewhat like the parts of a cane mill through which the sugar cane passes in expressing the juice, except that their surface is corrugated, the elevations of the one fitting into the depressions of the other.

The œsophagus is not more than 1 millimeter in diameter. The insect begins the process of separating the fibers of the tree by means of its chisel-like teeth. The rapidity with which a beetle can work is shown by the fact that within half an hour it will have entered a fourth of its own length into the plant tissue, and when once it is enabled to brace its strong-spined legs against the walls of the burrow its progress is accelerated. The heart of the tree is its objective point.

On Plate V are shown successive layers of leaf petioles at the heart with the burrow of an adult which finally reached the center. Figs. 1, 2, 3, and 4, respectively, show the pieces from outside, inward.

An examination of the fibers as soon as they are cut by the beetle demonstrates them to be almost dry, which renders it more probable that the purpose for which they are taken into the mandibles is solely to extract their juice, after which they are expelled from the mouth. Plate V, fig. 4, shows the heart of the coconut tree with a beetle at work. The bits of tissue which have been chiseled off can plainly be seen. In less than ten minutes the insect had burrowed into the soft substance for a distance of 10 millimeters. Plate III, fig. 2, shows the initial work of a beetle in a leaf petiole.

The beetles fly only at night; in the daytime they are readily found in their burrows. Their wings are quite large and the wing muscles in the thorax are strong and adapted for the flight of such heavy, unwieldy insects. In the interspaces between the intestines and the reproductive organs the abdomen is filled with air sacs and tracheæ.

Extent and character of damage done.—It is rare to find a single coconut tree anywhere in the Philippines which does not show one or more evidences of attack by this beetle. It is the pest most frequently reported by farmers and coconut growers, and in hundreds of trees which I have personally examined large holes in the trunk, distorted leaf stems, or ragged leaves demonstrate the character of its work. The insect larva or the adult, in its work inside the tree, frequently cuts off the tip of the embryo leaf or the tips of the leaflets on one or both sides of the midrib, so that when the leaf finally grows it appears as if it had been trimmed with a pair of shears or as if a triangle had been cut from one or both sides. The fibers severed by the insect protrude from its burrow, giving the latter a ragged appearance. During the daytime the beetles are frequently encountered in very old holes, into which they evidently have gone for the purpose of hiding. They have never been seen further to excavate these old cavities. The openings which are made serve to allow rain water to enter the tree, where it causes a most rapid decay of the interior, and they also serve as an entrance for other insects quite as destructive as the coconut beetle.

Distribution.—*Oryctes rhinoceros* L. is probably tropicopolitan, being found in Honduras, India, Ceylon, Java, the Philippines, Celebes, Borneo

and Sumatra, and recorded as coming from Africa. It undoubtedly thrives well wherever the coconut is grown.

Dr. Köningsberger, of Buitenzorg, Java, says: "The well-known coconut beetle *O. rhinoceros* L. is one of the most terrible enemies of coconut culture." And if this be the case in Java, where cultural methods have been in vogue for so many years, it is probably much truer in the Philippines.

Preventive and remedial measures.—The question of controlling the ravages of this insect is a difficult one. It would appear that trees growing to such a height as the coconut and having so few parts would not be seriously affected by a rank growth of weeds or underbrush or by a lack of cleanliness in their surroundings, but this is certainly not the case. It has already been stated that the larvæ of the coconut beetle grow in manure and rotting vegetable heaps and also thrive in rotten or rotting coconut trees, their abundance appearing to be in direct ratio to the degree of decay which the tree has attained. Mr. Schultze has taken as many as a hundred larvæ of all sizes from the decayed shell of a coconut trunk. I have invariably found them in great abundance in such situations. It is obvious that these sources of infection for healthy trees must be removed. The first thing to do in coming into possession of a coconut grove or in planting a new one is thoroughly to clean the ground. All manure heaps, rubbish, rotting or fallen trees should be removed and destroyed at once. The manure should be scattered where it will serve the best purpose as a fertilizer, and in such a manner as to make it impossible for the grubs to find lodgment in it. Rubbish heaps and decayed trunks, if fallen, should be burned; if standing, should be cut down and burned. The residue can easily be returned to the soil as fertilizer.

Growers should not remove the dead leaves from their trees to such an extent as to leave the young and still tender petioles or leaf stems entirely exposed, thus inviting attack by the adult beetles. These leaf stems have a thorough natural protection by being wrapped in a woven fiber, the old stems remaining upon the tree until the new ones are fully grown. When the living leaf stems are cut off a foot or two from their union with the trunk, the sap running out offers an attraction to beetles which might otherwise not attack the tree. Blanford discusses this danger as follows:[2]

"The trees should be left, as far as possible, in the natural state, and unnecessary trimming either of fronds or of the fiber avoided. It may be necessary to tie up the older fronds, and, if they must be removed, the stalk should be cut through sufficiently far from the stem to leave the sheathing base intact. It may be advisable to tar the cut stump if it is found to attract beetles. The value of leaving the trees alone is shown by a passage in Ferguson's *All about the Coco-nut Palm*, which is also quoted by Ridley: 'Scores of instances might be recorded

[2] *Kew Bulletin* (1893), 73, 46.

where, till the trees were come into bearing, a red beetle was never seen, but no sooner was the land cleared and the trees trimmed than it made its appearance and became very destructive. On one property the trimming system had been carried on for years, till, indeed, more than one-third of the original plants perished, before the estate was 10 years old, and they were going at the rate of three trees weekly. The work of trimming was stopped for the reasons offered above; the loss of the trees continued for some time afterwards, but at the end of six months it had entirely ceased. On another property beetle men had been employed for ten years, and trees were being constantly lost; from the day that the beetlers were discontinued two trees perished within a month, and not another was lost in the subsequent seven years.' And W. B. L. writes in the *Tropical Agriculturist* to the same effect: 'The red beetle (*Rhynchophorus ferrugineus*) can not penetrate the leaf imbrication, and, when the older ones decay in the course of nature, the stem has become too hard for its operations. A tree here and there may be lost from an accidental wound or from some defect in the fitting of the leaf sheaths, but it is only where the good taste of the planter has impelled him to trim the leaves that any serious damage has been done to a field. All the leaves should be left on the tree till nature disposes of them at her own time and in her own way. Nothing that can be done to a coconut tree above ground can be anything but injurious.'

"All wounds, whether made by accident or by insects, on the soft part of the stem, leaf sheaths, or spike should be at once dressed with a dab of tar mixed with fine sand. Holes should be probed with a "beetle spear" or hooked wire to extract insects which may have caused them and then plugged with a tuft of fiber or dry grass dipped in tar."

Obviously, no tree should be condemned until a careful and thorough inspection makes it certain that it is beyond hope of recovery and that it can bear no more fruit. It has been suggested by certain writers that a good plan is to cut such felled trees open, leaving them on the ground to attract beetles which would otherwise fly to the healthy trees; but I am of the opinion that the less material of this kind there is in the orchard the less is the liability of attack incurred by the bearing individuals. If there are no wounds or vulnerable spots in the trees themselves, and if nothing remains on the premises to attract this beetle and others, the less will be the danger. The dead leaves should be allowed to fall in the natural course of growth and care should be taken not to mutilate the trees. However, in most instances the beetles already have invaded the plantations and the serious problem is how to rid these places of them and to prevent their reëntrance. Of course, frequent inspections are necessary, so as to detect invasions at the earliest possible moment, because, as the coconut beetles hide in their burrows during the day, it is comparatively easy to destroy them if they are noticed in time. When they are discovered, a long, hooked steel wire can be thrust into the burrow, given a half turn to engage the insect upon its point, and then drawn out. This operation requires some practice, as the beetle is well armored with a smooth coat and has few projections upon the body. Dr. Könings-berger suggests crushing the insect and leaving it in the burrow as an obstacle to the entrance of others; but this is not to be recommended in

the Philippines, because the dead material would be sure to attract ants, which in turn would draw other insects, such as white ants (*anay*) and serious complications might arise. When the insect has been killed and removed, it is essential to plug the hole with some substance which will prevent further attacks at that point. For this purpose various substances have been recommended, for example: tar and fine sand, plaster and sand, clay and tar, or, in place of clay, plaster or cement. This mixture should be forced into the holes as far as possible, because it then will act as a deterrent to the decay caused by the entrance of moisture subsequent to attacks of the beetle, while effectually closing an avenue of entrance for others. Another remedy is to use a paste of Paris green and flour, mixed with 10 or 12 gallons of water, and sprayed into the crown of the tree. This method would offer difficulties when tall trees are to be dealt with, owing to the impracticability of getting the spray to the right places. The Filipinos use various remedies, such as sand and coarse salt, which they place in the crown of the tree. They state that the sand gets between the articulations of the head and thorax of the beetle, where the constant friction sets up an irritation which eventually punctures the soft tissues, after which the insect dies. This may be true. There is a constant movement of the head and of the thorax, while the beetle is working its way into the tree, and although the articulations are protected, as explained above, by a fringe of closely fitting bristles, it is possible that fine sand might enter as suggested and thus seriously handicap the beetle in its boring operations, if not eventually killing it.

I have been assured by a gentleman who is one of the most successful farmers in the Islands that natives on his plantation pour urine into the crown of the affected coconut trees and that this method effectually rids them of the pest. This is certainly not impossible.

ASIATIC PALM WEEVIL.

Rhynchophorus ferrugineus Fabr.

"It has been observed that coconut palms, the green leaves, blossoms, and fruits of which appear in perfect condition, fall to the ground, without having any signs of decay, as though struck by a hurricane. In such instances it has been noted that (the trees) from the roots up to a meter in height, are completely undermined, the interior pulverized like sawdust and filled with nests of these worms, which have gained entrance through the roots and gnawed their way upward, deriving maintenance from the trunk." [3]

The gravity of the attacks of the Asiatic palm weevil is well summed up in the foregoing extract, for, while the condition referred to is not generally reported from all parts of the Islands, there is every reason to

[3] Extract of translation of a communication from Señor Vicente P. Reyes, of Santa Cruz, Laguna, with reference to *R. ferrugineus* Fabr.

believe that in general the depredations of the beetle are no less serious in their ultimate effects than in the cases reported from the Provinces of Laguna and Tayabas, in which regions Señor Reyes has seen the damage to which he has made reference. I visited Magdalena, Province of Laguna, which lies in a fairly productive coconut region, and have found conditions closely resembling those set forth above, except that only a very few trees has actually fallen. In most instances where this had occurred the stumps had been cut off rather close to the ground during the previous year, and hence we found little material at hand upon which to work. However, we were convinced that the menace to coconut growing from this insect is fully as serious as, if not more so than, that occasioned by the attacks of *Oryctes rhinoceros* L.

This weevil enters the tree through the smallest wounds, leaving no external trace of its work, so that all its ravages are committed where not suspected; hence it is an extremely difficult enemy to combat.

The Asiatic palm weevil belongs to a group the members of which are, almost without exception, destructive to vegetable substances, either living or in the form of stored products, such as grains, beans, pease, and nuts. The beetle under discussion is one of the largest of its kind. The rice weevil is not more than 5 to 6 millimeters long; the corn weevil, *Calandra oryza* Linn., is slightly larger; the boll weevil, *Anthonomus grandis* Boh., which is at present proving so serious a menace to cotton growing in the United States, measures about 5 millimeters in length; the plum curculio, *Conotrachelus nenuphar* Hbst., a weevil, is 6 millimeters long; while there is another species attacking the coconut which measures 13 millimeters. The Asiatic palm weevil measures 35 millimeters in length. The form is strikingly characteristic in all individuals of this group. The most prominent features are an oblong, oval-shaped body and a long, slender, curved snout or bill, to which are attached the antennæ, either near the base or the tip. The colors vary from black to light brown or red, but are usually obscure.

The larvæ are legless, with a head of chitinous or horny structure, usually darker than the body and having two strong mandibles well adapted to gnawing the hardest vegetable substances. The Asiatic palm weevil has never been seen to make a primary attack upon the hard wood of the coconut; wherever it has been observed, it has utilized the holes made by *Oryctes*, wounds carelessly made around the base of the tree, or the steps cut into the sides of it by the tuba gatherers. Wherever the hard bark is broken and the softer parts beneath exposed, excellent places for the laying of the eggs are found and the beetle often makes a hole 10 millimeters deep before depositing them. The character of the hole and the tracks of the larvæ after hatching are shown diagrammatically in Plate VI, fig. 4. In laying their eggs in the burrows made by *Oryctes*, the palm weevils undoubtedly make no appreciable hole, simply

forcing the egg a short distance into the soft material in which the burrow lies.

Egg.—The egg is slender, 2.4 millimeters long and 0.6 millimeter wide at the middle, slightly more pointed at one end than at the other, and of a very light ocher. The shell appears perfectly smooth and shiny, but when examined under the microscope the surface is seen to be finely reticulated. (Pl. VI, figs. 1 and 1 *b*.)

Larva.—The larva does not vary essentially in general characteristics from the time of hatching until it is fully developed. Full grown, it measures from 35 to 55 millimeters in length and from 18 to 22 millimeters in diameter. The greatest diameter is slightly behind the middle. The hinder part of the body forms a concavo-convex extension, a blunt spoon or scoop-shaped organ.

The *head* is from 10 to 12 millimeters long and 7.5 to 8 millimeters wide. Seen from above, it is of a regular oval outline. It is of a dark-chestnut brown, with a slight reddish tinge, and with a lighter median and 2 submedian narrow stripes marking the sutures. (Pl. VI, fig. 2.)

The space around the mouth parts and the latter themselves are of a dark brown, with the exception of the upper lip and clypeus, which are lighter. The triangular portion of the head, immediately above the mouth, is transversely rugose, with a longitudinal furrow on each side of its middle. The remainder, including the cheeks and occiput, is engraved with very shallow reticulations, giving the appearance of a piece of alligator skin in miniature. The smooth, dark-brown, subtriangular, rather blunt mandibles are exposed on each side of the mouth; the upper lip, or labrum, lying between them, and reaching nearly to their tips, is provided with numerous bristle-like hairs. The larva has no antennæ, but the maxillary and labial palpi are well developed and doubtless serve as feelers. It has no eyes. The underlip, or labium, is subtriangular and rather small, but quite fleshy; the palpi project conspicuously from each side of its tip. (Pl. VI, fig. 3.) It is supposed that the surface of all these organs is highly sensitive and that the insect can tell desirable food by touch. The head shows erect hairs placed at regular intervals, 5 on each side of the top and 3 back of each mandible. Portions of the front of the head, and the mandibles, appear as if having been rubbed off by friction with the substance in which the larva lives, so that these parts have a dull, almost black or *matte* appearance.

The *body* is composed of a series of 13 rings very much folded and wrinkled, the surface being of a smooth, velvety texture, except in certain spots, which are decidedly shiny. On the back of the first segment appear 2 transverse, oblong patches of a darker color than the remainder of the body, with a surface similar to that of the head and serving as a protection to the animal in its movements in the small galleries in which it works. There are similar lighter areas at the sides of the first 3, or thoracic, segments, which are somewhat swollen and serve in lieu of legs. Scattered over the entire surface are tiny, circular or irregular shiny areas, from each of which arises a small curved bristle. On next to the last segment of the body, dorsally situated, are also irregular shiny patches, each with 6 bristles. The last segment has the upper surface concave, and the lower convex; the posterior margin, which is slightly darker than the rest and smooth and shiny, is flattened out and has four prominences, from each of which project two rather long bristles. The spots from which they project are lighter in color. The wrinkles of the body are nearly symmetrical. (Pl. VII, fig. 1.) The body curves downward, so that the back is very convex, while the underside is somewhat concave, except just back of the middle, where it is convex, then suddenly tapering to the tip.

This grub works its way forward through its burrows by a combination of twisting and undulating motions. In this it is aided by the tubercular enlargements on the thoracic segments. The hind end appears to offer no help in this respect. It can enter from any opening through which the head will pass. The bristles on the head serve as guides for the insect in passing through holes. When placed upon a level surface, the grub moves slightly sidewise, almost always upon its side, and can thus make fairly rapid progress.

The breathing apparatus in *R. ferrugineus* Fabr. consists of only two pairs of spiracles which are well developed, the others being almost rudimentary. Each of the first pair is situated laterally on the first thoracic segment, and twice its own length below the shiny, transverse, shield-like areas, and the second pair just above the spoon-shaped excavation of the thirteenth segment of the body. The latter two are one and one-half times as large as the first two, and their openings are nearly vertical, diverging slightly below. The other segments of the body show the spiracles only when examined under a strong lens; these are nonfunctional, or at most only very slightly so.

The galleries of this grub run obliquely through the large swollen part of the tree near the roots. (Pl. VIII, fig. 3.) The specimen here depicted was full of grubs of all sizes and contained one or two pupæ as well. Adult beetles were also found in considerable numbers. The grubs have been encountered in the crown of the tree in numbers varying from 15 to 20, where they work side by side with those of the rhinoceros beetle, and it is very difficult to distinguish the galleries of the one from those of the other. Plate III, fig. 1, shows a longitudinal section of the crown of a tree which has been eaten out in the form of an inverted cone by the larvæ of the rhinoceros beetle and the palm weevil in company. Plate VIII, fig. 3, shows the work of the weevil in the lower part of the tree very near the roots, some of which are seen at the lower right-hand corner. It will be noted that the galleries run obliquely, which shows that the grubs work inward and upward from the outside of the tree. In this case the eggs were evidently laid in wounds in the root region, on the left, and the grubs worked their way toward the center where the full-sized galleries are seen.

Pupa.—When the larva of the palm weevil has attained its full size, it ceases feeding and evacuates the alimentary canal, thus causing a shrinkage of one-third in its size immediately before making its cocoon. This is elliptical in outline, from 8 to 12 centimeters long and 5 to 6 millimeters in diameter, and composed of the long tough fibers of the coconut trunk wound as shown by Plate VII, fig. 3. It is closely woven and thick, so that the pupa is well protected against dampness. The grub sheds its skin and takes the form shown by Plate VII, fig. 2. The pupa measures 35 to 40 millimeters in length and about 15 millimeters across its widest portion. The snout is doubled down on the breast; the antennæ, wings, and other organs of the beetle are plainly visible. The color is a uniform pale-ocher, the tips of the knees being darker. Rugose areas are situated on each side of the head, back of the eyes, on the upper part of the snout, on the outer fore and hind regions of the pronotum, on the ridges of the elytra, and on the dorsum of the abdomen. These areas are rather thickly

set with short, sharp spines, which aid the beetle in escaping from the pupal skin by holding the latter firmly in the cocoon. The spiracles, which on most of the abdominal segments in the larva were nearly obsolete, are very prominent in the pupa, a pair on each of the first 6 segments and a fainter one on the seventh being visible.

Up to the present it has been impossible to ascertain the length of time of either the larval or the pupal stage. The beetles begin their work in the trees at practically the same time as *Oryctes* and the adults are found together with those of the latter, so that the life periods in the larva and pupa of both are probably about the same, or from 18 to 24 months.

Adult.—Rhynchophorus ferrugineus Fabr. is an extremely variable insect, in its markings as well as in its size. Specimens have been obtained of from 25 to 35 millimeters in length, while the color varies from a true ferruginous, with certain black markings more or less regularly placed, to almost entirely black, with only traces of ferruginous. (Pl. VIII, fig. 1.)

***Rhynchophorus* sp.**

This species is very closely related to *R. ferrugineus* Fabr., if not identical with it, merely varying in general color and in having a broad, ferruginous, longitudinal band from the front to the hinder margin of the thorax. (Pl. VIII, fig. 2.)

The habits and the immature stages of this insect are similar to those of *R. ferrugineus* Fabr. These beetles are found indiscriminately in company on the same tree, and no differences are noted until the adults are compared.

Preventives and remedies.—The prevention of the first attack of the pest is essential. The adult male or female can not bore into the solid tissue as can that of the rhinoceros beetle, because the snout is small and the mandibles are relatively weak. For this reason the female seeks wounds or holes of any kind in the trunk of the tree to deposit her eggs. These wounds may have been caused by other insects, or they may be accidental. One of the chief injuries to the trunk of the tree is that caused by the gatherers of tuba, who make notches in it whereby they may be enabled to climb to the top. As these notches are of considerable size and depth, they offer excellent facilities for the beetles to enter and hide or lay their eggs. All such mutilation of coconut trees should certainly be prevented, even if it be necessary to construct bamboo ladders, securely fastened to the trees, as is done in some localities. There are frequently encountered in coconut plantations trees the bases of which seem to have been chopped with no apparent purpose in view. Of course, these offer an excellent opportunity for the beetles to begin their work. A good, healthy, vigorous, uninjured coconut tree is practically invulnerable to the attacks of the palm weevil.

If, in spite of all precautions, the weevils gain entrance to the tree, the work of combating them is exceedingly difficult. Frequently, when

they are in the softer, upper parts, it is possible to dig them out with a wire hook similar to the one mentioned as effective against the rhinoceros beetle larvæ. In every case where these or other larvæ are dug out of a burrow, this should at once be filled with a substance distasteful to the adult beetles. Great care is necessary in the work of extracting the larvæ, lest it should be carried to such an extent as to debilitate or kill the tree. If the weevil larvæ are located at or near the base of the tree, where it is almost impossible to dig them out, the only practical method is to stop all avenues of escape and then to remove the tree after it ceases to bear fruit. It has been suggested that infested trees be cut down, split lengthwise, and then left to attract beetles from the others. I am opposed to such a procedure, because it would surely attract fully as many insects from a distance as it would from the immediate neighborhood. Such a method would be advisable only if the other trees were in great danger from beetles already present in them. Plate II shows a coconut tree very badly infested by both the rhinoceros beetle and the Asiatic palm weevil. It will be seen that the entire interior has been eaten out and converted into a mass of débris, in which both the cocoons and the larvæ of these insects were found in great abundance. The tree had cased to bear, the growing point was gone, and there remained only a circle of older leaves, kept alive by the small flow of sap in the outer shell of the trunk. It is obvious that such a tree is a source of infection for a large area.

THE SHOT-HOLE COCONUT WEEVIL.

This destructive weevil has been found in Laguna Province in considerable numbers. I once felled a dead coconut tree, the trunk of which was completely pitted from top to bottom by the insects' exit holes, and Mr. Schultze found the larvæ and pupæ as well as the adults in a living tree. (Pl. X, figs. 1 to 5.)

Egg.—The egg has not been found. It is probably laid directly in the hard wood in small cavities made by the female, as the grub can work in any part of the trunk of a tree.

Larva.—The larva is a very pale-yellow, almost white grub, measuring 16.5 millimeters in length and 6 millimeters in diameter, resembling the larva of the palm weevil, except that the hinder end of the body is evenly rounded. The head is shiny and but slightly darker than the rest of the body, the region around the mouth and the mouth parts appearing dark-brown. A very thin, brown median line runs from the upper lip halfway to the back of the head. The spiracles are very small and almost black. The surface of the body is smooth and very much folded. A few bristle-like hairs are seen on the head. (See Pl. X, fig. 1, illustrating a full-grown larva.)

Pupa.—The pupa is 13.5 millimeters long and 6 millimeters in diameter and of the same color as the larva. The surface is smooth and shiny. On the head, thorax, and dorsum of the abdomen there may be seen a series of stout, brown bristles arising from brownish tubercles. The tip of the abdomen has a small, white tubercle on each side, from the point of which arises a small, black bristle. There is little difference between the size of the tubercle and that of the bristle.

The pupa rests in a cavity in the live wood, there being no attempt at forming a cocoon. (See Pl. X, fig. 3, giving a lateral view of the pupa.)

Adult (Pl. X, fig. 4.).—The adult both in form and in size appears very much like the willow weevil, *Cryptorrhynchus lapathi* Linn., of the United States, except that there are no tubercles on the thorax and wing covers. It measures 11 millimeters in length (exclusive of the bill, which normally is doubled under the body) and is 5.5 millimeters in its greatest breadth. (Pl. X, fig. 4.) It is of a dark-reddish brown, somewhat mottled with gray on the forward part of the thorax, which is very closely punctured. The head is globular and fits almost entirely into a cavity in the front of the thorax, so that, when seen from above, it has the form of a thin crescent. The eyes are black and somewhat oval, nearly meeting on the front of the head, the space between being one-fourth the width of the rostrum or bill. When the insect is at rest the antennæ, which are inserted on each side of the rostrum near its base, are completely hidden, being drawn within the cavity in which the head fits. They are geniculate, the apical part or flagellum being somewhat more than half the length of the bill, very slender at its base, and increasing in size toward the club-shaped apex, which has three segments very closely united. There are 12 segments in the antennæ, of which 11 are in the flagellum. (Pl. X, fig. 2.) The surface of the 3 apical ones is very pilose and of a sensitive nature. The rostrum is smooth, closely punctured, and slightly broadened at its apex. The mandibles are plainly visible, slightly darker than the rostrum, and uniting at their apices to form a triangle. The part immediately above the mandibles is covered with strong, light-brown bristles, pointing toward the tip of the rostrum. The thorax bears a longitudinal depression which is light-gray in color, owing to the scales on the surface, and which extends nearly to the posterior margin, where the depression becomes a ridge or carina one-sixth the length of the thorax.

The elytra reach nearly to the tip of the abdomen, are very rough, and are traversed longitudinally by nine rows of punctures forming very deep grooves; six of these extend to the apex of the wing covers, the others being interrupted or running into each other. The external (ninth) row terminates before the middle of the elytron. The posterior portion of the propluræ shows a decided depression, into which the front legs fit on each side.

The *legs* are moderately long and very stout, the fore pair being nearly a half longer than the other two. The rostrum reaches beyond the insertion of the first pair; there is a transverse carina of the mesosternum against which it rests. Two spines are situated on the under sides of the femora near their apices, the smaller nearer the apex, and those on the forelegs larger than those of the middle and hind pairs. The tibiæ are all of the same shape, each bearing a curved spine or tooth and 3 bristles at its apex; the latter are external.

The *tarsi* are 4-jointed, the fourth being very small and hidden between the pulvilli or pads. The tarsal claw is bifurcated, very long and slender. The tarsi are covered with long, blunt, silvery-white scale-like hairs.

The exposed part of the pygidium, or hinder segment of the abdomen, is bluntly, almost emarginately rounded; the apical half is covered with golden-brown bristles lying flat. The last ventral abdominal segment is hairy apically. The beetles appear to present no external sexual characters.

Remedies and preventives.—Doubtless these insects would be susceptible to the same general treatment as that given to the Asiatic palm weevil, although too little is known of their habits to be certain. They have been found in all stages, generally in diseased trees or in those debilitated by the attacks of other insects, and hence should not form a serious menace.

THE BONNOGA WEEVIL.

Cyrtotrachelus sp.

This weevil lives principally in the trunks of the betel palm, *Areca catechu* Linn., where it does great damage, but inasmuch as it has also been found in considerable numbers in coconut trees, it is here described as far as its habits and appearance are concerned. In addition to the larvæ of the rhinoceros beetle and the cocoons and grubs of the palm weevil, one frequently encounters in decaying betel palms or coconut trees of 6 to 8 years, other smaller cocoons not more than 35 millimeters long and 15 millimeters in diameter. (Pl. VII, fig. 4.) These are composed of a more finely comminuted fiber than those of the palm weevil, and upon opening appear to contain a dwarfed example of the pupa of the Asiatic weevil. However, this pupa differs in many respects from that of *Rhynchophorus ferrugineus* Fabr., and the frequent finding of beautifully marked weevils of very small size convinces one that these cocoons and pupæ belong to the former.

Egg.—No eggs have been encountered, and attempts at confining the adults for the deposition of eggs under conditions as nearly natural as possible have failed.

Larva.—The full-grown larva is nearly of the same size as the preceding one. However, in form it is more like that of the Asiatic palm weevil and is probably somewhat closely related to it. The color is a light-ocher yellow. The head is very much darker, and the mouth parts are dark-brown.

The length is 20 millimeters and the diameter 6 millimeters near the rear third of the body, the form being strikingly like that of the Asiatic weevil in this particular. The head projects forward and is smooth and shiny, with but few hairs scattered over it.

The spiracles on the first thoracic segment are larger than any of those on the other ones of the body, with the exception of the last abdominal segment, on which they are well developed and placed on the posterior aspect. The apex of the last segment is somewhat flattened and its hinder margin is prolonged into 4 rather obscure tubercles, from each of which arise 2 bristle-like hairs pointing posteriorly and slightly downward. Certain areas on the skin of the entire body, except the head, are rough or very minutely shagreened, single isolated hairs arising from some of them.

The mouth is minute, but the upper and lower lips and the mandibles are well developed, the latter being black. The palpi are prominent. As with the larvæ of *Rhynchophorus ferrugineus* Fabr., there are no evidences of external eyes or ocelli. (Pl. XI, fig. 1.)

Pupa.—The pupa is illustrated by fig. 3 on Plate XI. Its length is 13 millimeters and its greatest diameter 6 millimeters. It is of a whitish-ocher color, certain of the tubercles being a darker ocherous. On the front of the head, just above the point where the eyes would appear in the adult, there are 2 prominent corrugated tubercles, each with a single bristle; anterior to these are 2 smaller ones with bristles; and on the snout or rostrum above the antennæ are 2 more. On the front margin of the thorax are 2 other tubercles, smaller than the largest on the head; on the anterior half of the thorax toward the sides is another pair; and near the posterior margin are 2 others slightly larger; all of these are provided

with a single bristle. The first 6 dorsal abdominal segments are very sharply defined and bear on each side of the middle a transverse group of 3 tubercles with bristles; outside of these is a single one.

The spiracles are plainly seen at the latero-dorsal angle of each segment. The pygidial segment is curved downward and at its middle there is a transverse line of 8 tubercles with bristles, slightly separated on the median line. The apical part of this segment has a median, transversely corrugated carina. On the extremity of the last ventral segment, on each side, there is a concentrically corrugated tubercle, from which 2 yellowish ochraceous bristles arise.

Each femur, on the outer part of its apex, has a single tuberculated spine, darker than the surface. The pupæ are very active when taken from their cocoons, wriggling continuously if held in the hand.

A peculiar large, button-shaped spiracle may be seen on each side just behind and a little below the prothoracic shield.

Adult.—This insect is graceful in form and very delicate in color. A black and white drawing, such as fig. 4 on Plate XI, of course can not show its coloring, which is one of its most striking features. The profile is shown by Plate XI, fig. 6.

The length is 17 millimeters from the tip of the snout to the tip of the abdomen, and the diameter about 4.75 millimeters. Seen from above, it measures 13 millimeters in length. Its color is a combination of ocher, reddish-ocher, and dark-brown, or black, in which reddish-ocher predominates.

The *head*, exclusive of the snout, it globular, and smooth above and below, with a few scattering shallow punctures at the side. Its color is reddish-ocher. The eyes are jet-black and broadly crescent-shaped, nearly uniting at the upper and under sides of the head; their exterior outline, when viewed directly from in front, is almost a perfect circle. The rostrum is cylindrical and strongly curved downward, the basal third being twice the diameter of the rest and covered with circular light-gray spots, from each of which arises a tiny, dark-brown tubercular spine. The apical two-thirds is smooth and finely punctured longitudinally. The tip is slightly swollen laterally and of a darker color, as are the mouth parts. The mandibles are black and glossy, and tridentate, the teeth of one fitting into the interstices of the other. The narrow transverse labrum, with the anterior margin rounded, is scarcely visible. The antennæ, apparently composed of 8 joints, of which the first is equal in length to the others combined, are placed in short deep furrows on each side of the snout a little less than one-fourth the distance from its base. The upper edge of this furrow projects at its middle and somewhat overlaps the articulation of the first antennal joint. The last joint is greatly swollen, being twice the diameter of the preceding one, and is securiform. The length of the antennæ is equal to that of the rostrum. The third joint is chalice-shaped and one-half longer than the second one, which is inserted on the inner apical portion of the first. The prothorax is subconical, three-fourths as wide as the elytra, perfectly truncate at its anterior margin, and slightly rounded posteriorly. A narrow collar extends around its entire anterior margin, the sides of which are subparallel. The sides of the thorax are rounded, and their surface is smooth, dull, and sparsely punctured, the punctures toward the sides bearing small tubercles or spines. These punctures are also found on the underpart of the pro- and mesothorax, the metathorax, the ventral surface of the abdomen, the pygidium, the femora and the tibiæ of all the legs.

Thoracic markings occur as follows: A lancet-shaped, light-ochraceous or buff median mark, extending from a point behind the anterior margin one-fifth the length of the thorax, to the posterior margin; on each side of this a wide black line, broadening perceptibly at the posterior margin, meeting in front of the

median mark and extending to the anterior margin; outside the black lines, on each side, a reddish-ochraceous one, twice as broad as the median and not extending to the anterior margin; external to these on each side, a broad pale-ochraceous or buff line, mixed with reddish-ocher at its outer edge and running imperceptibly into the extreme outer longitudinal black lines on the sides of the thorax. The black lines are irrorated with buff and have buff punctures anteriorly.

The scutellum, between the bases of the elytra, is lancet-shaped, black, and shiny.

The *elytra* have a ground color of reddish-ocher, with the following longitudinal markings: A series of 9 punctured lines, 5 of which reach the apex; a buff line on the sutural margin and a similar, although redder, space between the fourth and fifth and the sixth and seventh punctured lines. Each space is interrupted several times before it finally meets with the others near the apex of the elytron. The elytra bear near the base ‖ and ‖ and on the apical half ⌜⌝ on the left and right, respectively. There is also a broad, black, uninterrupted band on the external margin, confluent with a similar one on the prothorax. In the male these black characters are more or less confluent. The apices of the wing covers are emarginate at the suture; the pygidium is truncate, with the sides gradually converging; the median portion of the ventral surface of the thorax and abdomen is black and glossy, with numerous spine-bearing punctures; the posterior margins of the meso- and metathorax are deeply notched; the fore coxæ or first leg joints are almost contiguous, the interspace having a transverse suture. An elytron of the female is shown by Plate XI, fig. 5.

The *legs* are stout and moderately long; the femora are slightly swollen at their apices; the tibiæ of the middle legs are somewhat shorter than those of the fore and hind ones, and all are longitudinally ribbed with spine-bearing tubercles of minute size. The apices of all tibiæ bear a large tooth and two stout bristles. The 4-jointed tarsi are covered sparsely above and densely beneath with golden-brown hairs. The bidentate claws are long and graceful. These beetles have no constant external evidences of sex differentiation. (See Pl. XI, fig. 2, showing hind legs.)

Remedies and preventives.—The same preventive measures and remedies apply in combating this insect as are recommended in the case of the Asiatic palm weevil. The damage done by them is not by any means so extensive as that due to the other insect, but, nevertheless, it should, if possible, be prevented or stopped, as the tree is finally killed by the summation of the attacks of the various insects which it harbors.

THE FOUR-SPOTTED COCONUT WEEVIL.

The length of this beetle, exclusive of the snout, is 5 millimeters, and the width is 1.5 millimeters. It was found in the dead or decayed heart or the undeveloped leaves of a small 3-year-old coconut tree during a search for the rhinoceros beetle. It attacks only dead trees of a very small size and is met with only in coconuts. In addition to the adult beetles, the larvæ and the pupæ were secured in numbers. Plate IX, figs. 1 and 2, shows the exit holes of the adults and the work of the larvæ in the interior of the tree.

Egg.—The egg of this species is not known. It is probable that the beetles deposit their eggs on the sticky sides of their galleries in the trees; though close search failed to reveal them; but, as these places are also occupied by many

other refuse-destroying insects and mites, it is probable that few of the shells would remain after the young grubs had emerged. Doubtless some of the mites feed upon the eggs themselves and in this way serve to limit the number of the beetles.

Larva.—The larva is of about the same general shape as that of the shot-hole coconut weevil, but it is more slender in proportion to its length. The length is 7 millimeters and the width 2.5 millimeters. The color is a pale-cream, the head being somewhat darker and the mouth parts dark-brown. The head bears numerous scattered, golden-brown bristles. The posterior margin of the labium is rounded, with a sharp angle at the median line. The ventral surface of the first thoracic segment is microscopically and densely spinose-tuberculated, as are also certain transverse areas on the ventrum and dorsum of the middle abdominal segments. The spiracles are extremely minute, somewhat slender, and pyriform, with lines radiating from the central slit to the margin. The upper, posterior surface of the last abdominal segment is slightly excavated, with 10 rather fine bristles; it is of a golden-brown on its margin. The larvæ feed in well-defined burrows or galleries slightly isolated from each other.

Pupa.—The length of the pupa is 5.5 millimeters and the width at the middle 1.75 millimeters. It is cream-colored and in general shape like the pupa of the shot-hole coconut weevil. Golden-brown spinose hairs are arranged as follows: Two pairs, very small, on the rostrum above the antennæ, 2 larger ones in front of the eyes, 2 still larger ones on the top of the head back of the eyes, 8 pairs symmetrically on the prothorax, 7 on the meso- and metathorax, respectively, 2 on each abdominal segment from the first to the sixth, 1 on the seventh, 1 at the base of the pygydium, and 1 on the ventral apical margin of the last abdominal segment, pointing downward. Each femur is provided at the outer apical angle with a single erect spinose hair. The spiracles are hardly visible.

Adult.—The general color of the beetle is dark-brown, with rufous patches. The head is globular and strongly punctured. The eyes are black and broadly crescent-shaped, contiguous beneath the head, but separated above by a narrow shallow sulcus at the base of the rostrum. The rostrum is slender, subcylindrical, slightly swollen laterally at the base above the insertion of the antennæ, and coarsely punctured, each of these punctures as well as all others upon the surface of the body containing a single club-shaped hair or bristle. (Pl. X, fig. 6.) The mouth parts are extremely minute. The mandibles are tridentate, and when closed are almost completely hidden within the mouth cavity. A narrow longitudinal sulcus is situated on each side of the mentum, into which fit the maxillary palpi. The antennæ are 8-jointed and of the same length as the rostrum; the first joint is slightly shorter than the other seven, the last is double the diameter of the preceding one and its distal half is silvery-pubescent with sensitive hairs.

The thorax is truncately conical and its anterior and posterior margins straight and parallel, the former having a narrow, smooth collar, back of which are numerous setigerous pits or punctures. It is coarsely and deeply punctured, with an indistinct rufous spot on each side. The scutellum is subtriangular and excavated at its middle. Each elytron is marked by 2 reddish-subquadrate spots, one at the base and the other beyond the middle, and is traversed longitudinally by 5 very finely punctured ridges or carinæ. Between every 2 carinæ there is a double row of very regular, coarse, deep punctures. The apex of each elytron is rather sharply rounded, the pygidium is subtriangular, and its sides and median line are carinated and rather densely setose, the setæ springing from fine punctures. It is very easily depressed; in some specimens it forms an angle of nearly 90° with the remainder of the abdomen. The legs are stout, moderately long, and the pairs about equidistant from each other and from the 2 extremities of the body, roughly dividing the latter into 4 subequal sections, if the rostrum

is excluded. The femora are swollen toward the apical angle and each has a slight constriction beneath and just in front of the apex. Each tibia has an apical claw and a tuft of pre-apical hairs beneath. The tarsi are all of equal length. The legs are conspicuously shiny in comparison with the rest of the body. Plate X, fig. 8, shows profile and fig. 7 an antenna of this beetle.

As is the case with nearly all weevils, these also feign death when annoyed. They conceal themselves under any available object and unless disturbed remain in one spot in their burrows for a long period.[4]

Remedies and preventives.—These beetles are found only in locations where others have preceded them and killed the trees; hence, they are not in any sense a menace to the healthy tree. Their description has here been given merely to call attention to all forms which may be encountered.

[4] It was hoped that the identifications of the shot-hole coconut weevil, the bonñgga weevil, and four-spotted coconut weevil would be received from Washington in time for insertion in this article, but as they have been delayed, it was thought best to publish the paper and give the identifications later.

[*Part II of this paper will treat of the insects which live upon the leaves of the coconut, including two species of Lepidoptera and certain species of Coccidæ, of which there are several that make their homes upon the coconut. There will also be given a bibliography of the insects of the coconut.*]

ILLUSTRATIONS.

PLATE I.

FIG. 1. Adults of *R. ferrugineus* Fabr. (About natural size.)
2. Head of larva. (X 5.)
3. Antenna. (X 12.)
4. Left mandible of female:
   (*a*) Profile and inner surface, showing condyle. (X 7.)
   (*b*) Interior view. (X 7.)
5. Profile of head and thorax of female. (X 1½.)
6. Profile of head and thorax of male. (X 1½.)

PLATE II.

Coconut showing results of attacks of *Oryctes rhinoceros* L. and *Rhynchophorus ferrugineus* Fabr.

PLATE III.

FIG. 1. Crown of coconut, showing inverted cone, in longitudinal section, eaten out by larvæ of *O. rhinoceros* and *R. ferrugineus* Fabr. (About one-seventh natural size.)
2. Adult of *O. rhinoceros* L. boring into petiole of coconut leaf. (One-half natural size.)

PLATE IV.

FIG. 1. Larva of *Oryctes rhinoceros* L.
2. Pupa of *Oryctes rhinoceros* L.
3. Adults, male and female, of *O. rhinoceros* L. (All about natural size.)

PLATE V.

FIGS. 1–4. Heart of coconut showing burrow made by adult of *O. rhinoceros*, and a female beetle working at center of tree. (One-half natural size.)

PLATE VI.

FIG. 1. Egg of *Rhynchophorus ferrugineus* Fabr.; magnified portion shown at 1 *b*.
2. Head of larva of same. (X 2½.)
3. Labium of larva of same. (X 3.)
4. Diagram of work of larvæ in base of coconut trunk, showing points of entrance, as at A.

PLATE VII.

FIG. 1. Larva of *Rhynchophorus ferrugineus* Fabr.
2. Pupa of *Rhynchophorus ferrugineus* Fabr.
3. Cocoon of *R. ferrugineus* Fabr., showing beetle partly emerged.
4. Cocoon of *Cyrtotrachelus* sp. (All about natural size.)

PLATE VIII.

FIG. 1. Adults of *R. ferrugineus* Fabr. (About natural size.)
2. Adults of *Rhynchophorus* sp. (About natural size.)
3. Work of larvæ of *R. ferrugineus* Fabr. in wood of coconut near roots. The larvæ entered from the lower left.

PLATE IX.

FIGS. 1, 2. Work of shot-hole coconut weevil in trunk of coconut, with exit holes of adults.

PLATE X. (Drawn by W. Schultze.)

FIG. 1. Larva of shot-hole coconut weevil. (X 3.)
2. Antenna of shot-hole coconut weevil. (X 3.)
3. Pupa of shot-hole coconut weevil.
4. Adult of shot-hole coconut weevil. (X 4.)
5. Adult of shot-hole coconut weevil, profile. (X 4.)
6. Adult of four-spotted coconut weevil. (X 8.)
7. Antenna of same. (X 36.)
8. Profile of same. (X 8.)

PLATE XI. (Drawn by W. Schultze.)

FIG. 1. Larva of *Cyrtotrachelus* sp. (X 5.)
2. Hind legs of adult *Cyrtotrachelus* sp. (X 10.)
3. Pupa of *Cyrtotrachelus* sp. (X 5.)
4. Adult male of *Cyrtotrachelus* sp. (X 5.)
5. Elytron of female of *Cyrtotrachelus* sp. (X 5.)
6. Profile of male of *Cyrtotrachelus* sp. (X 5.)

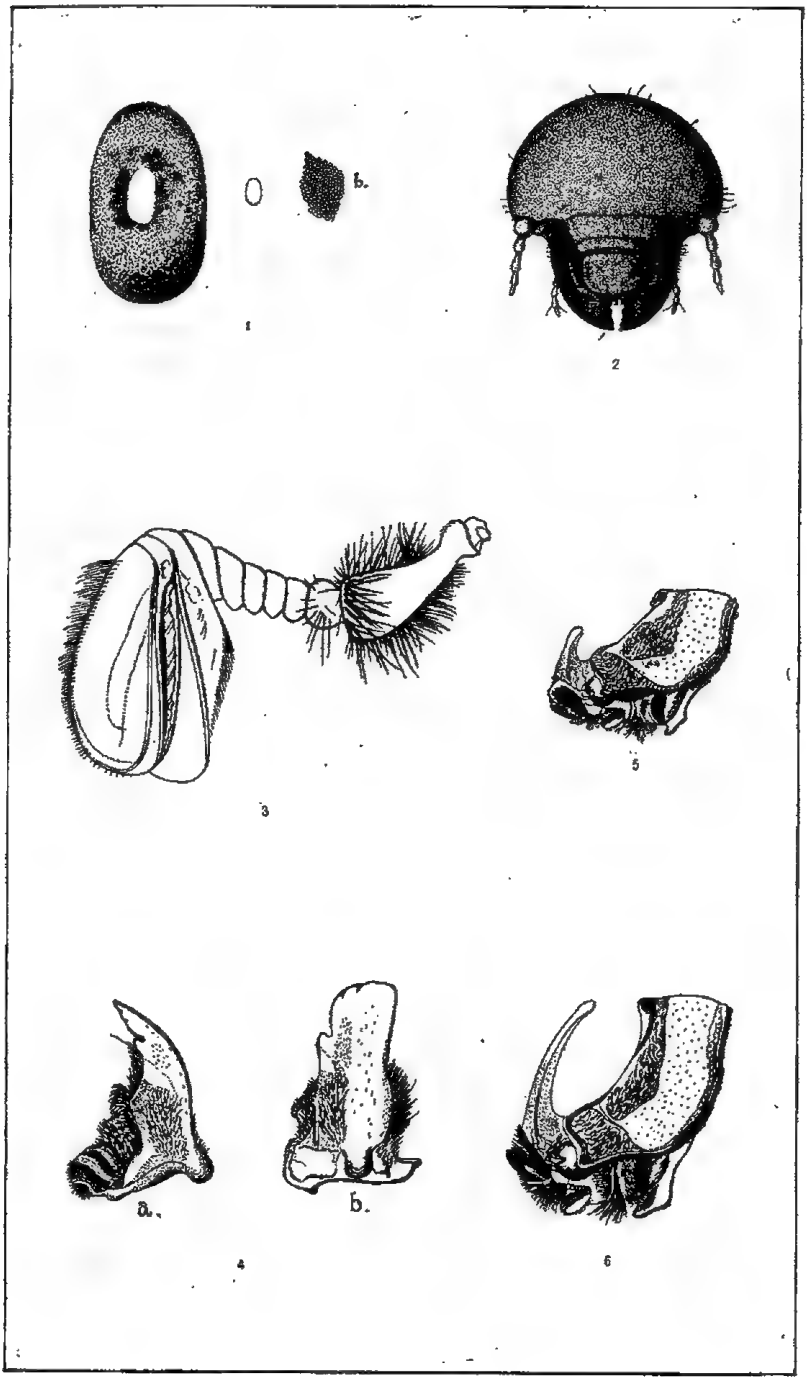


PLATE I.

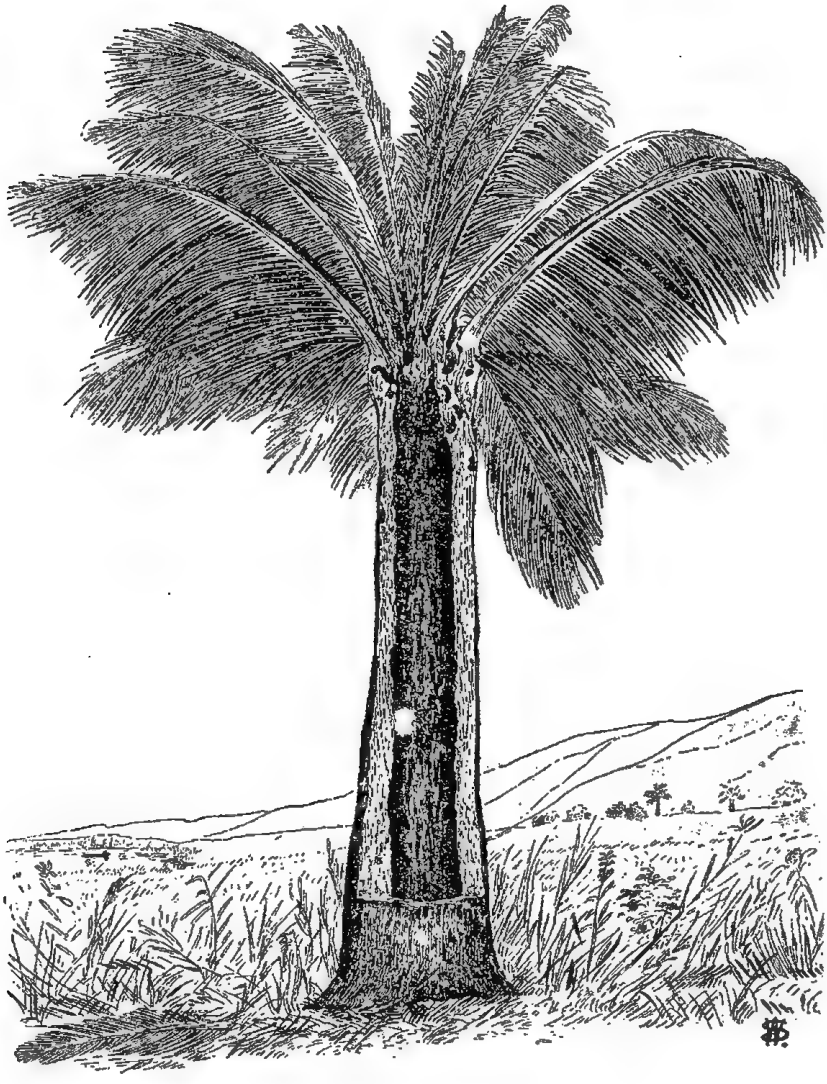


PLATE II.

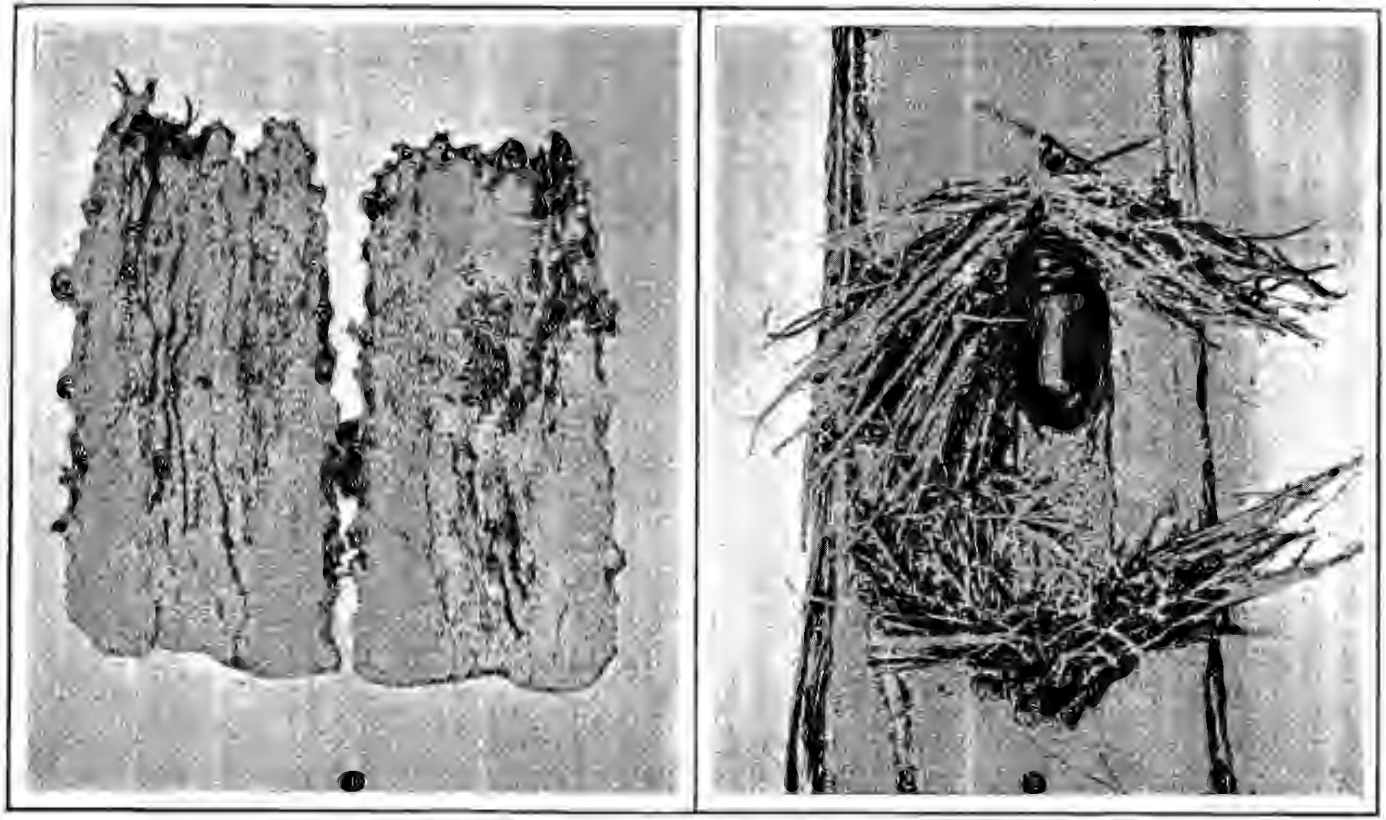
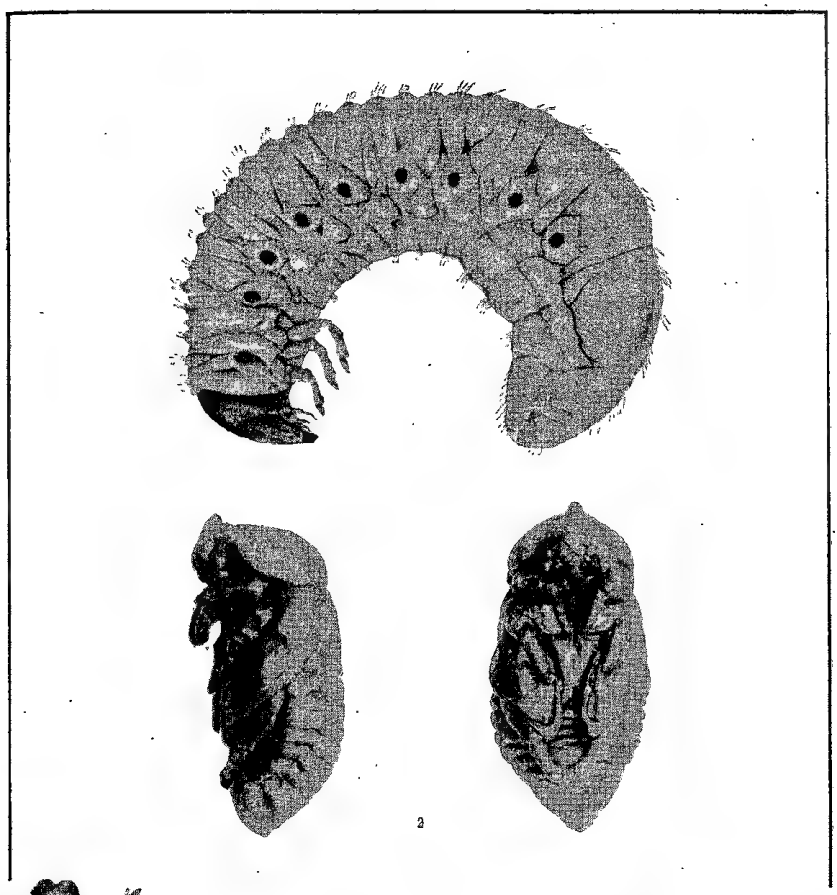
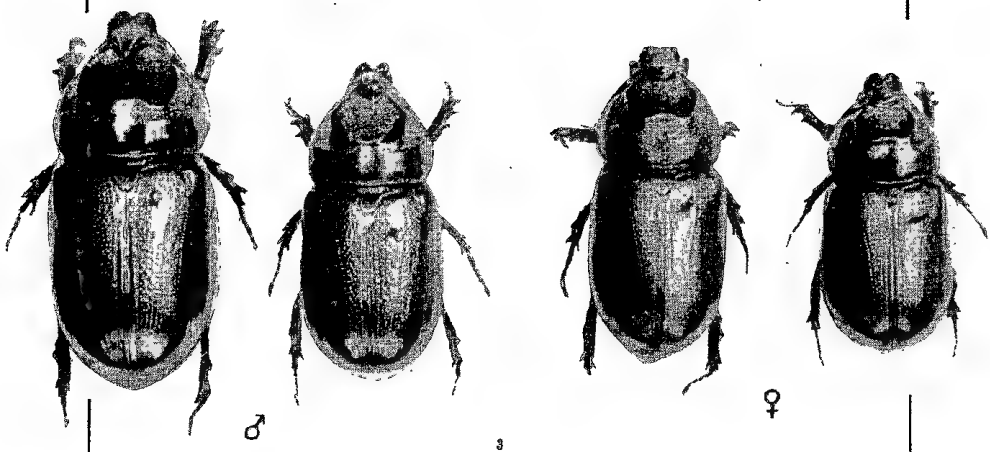


PLATE III.



2



3

PLATE IV.

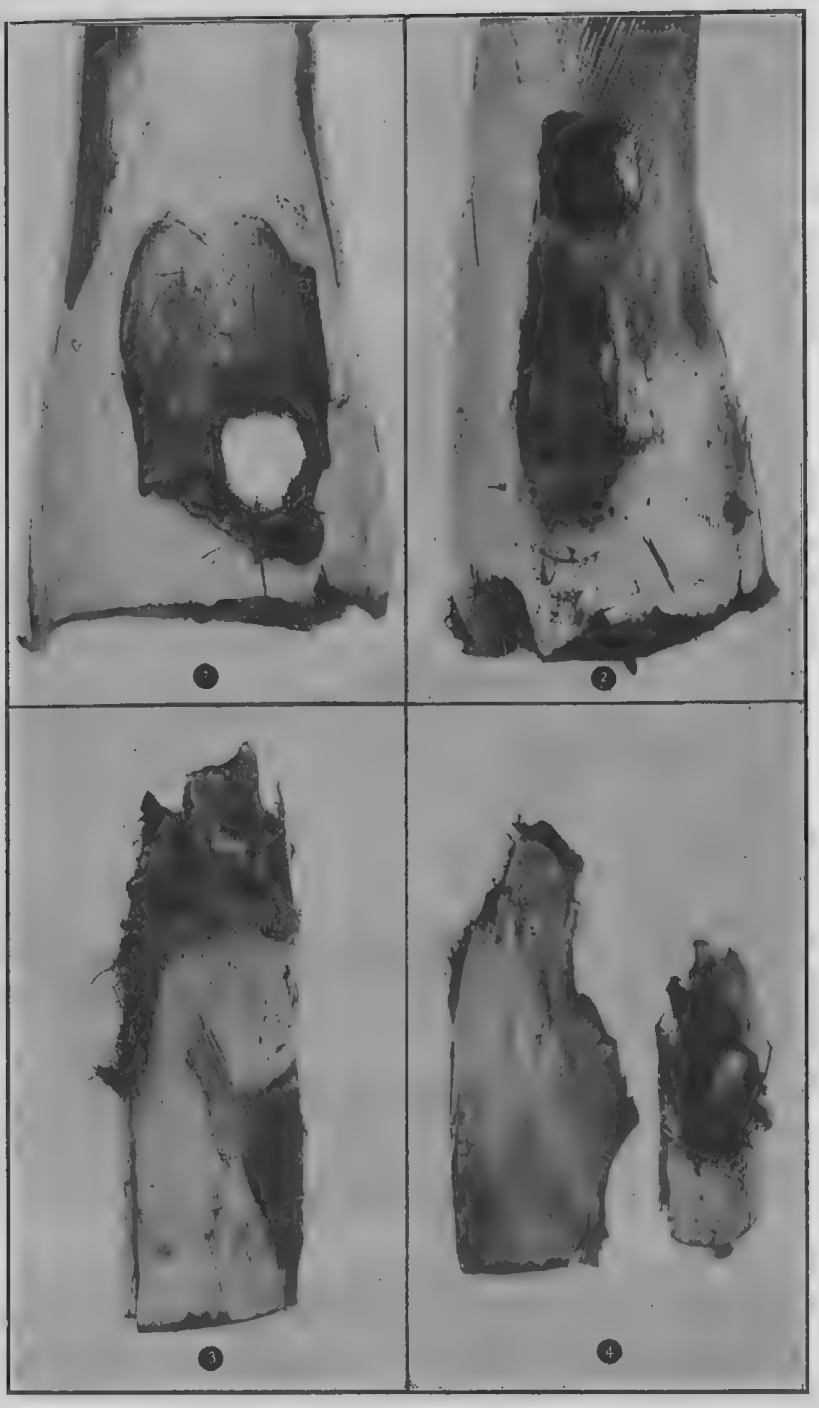


PLATE V.

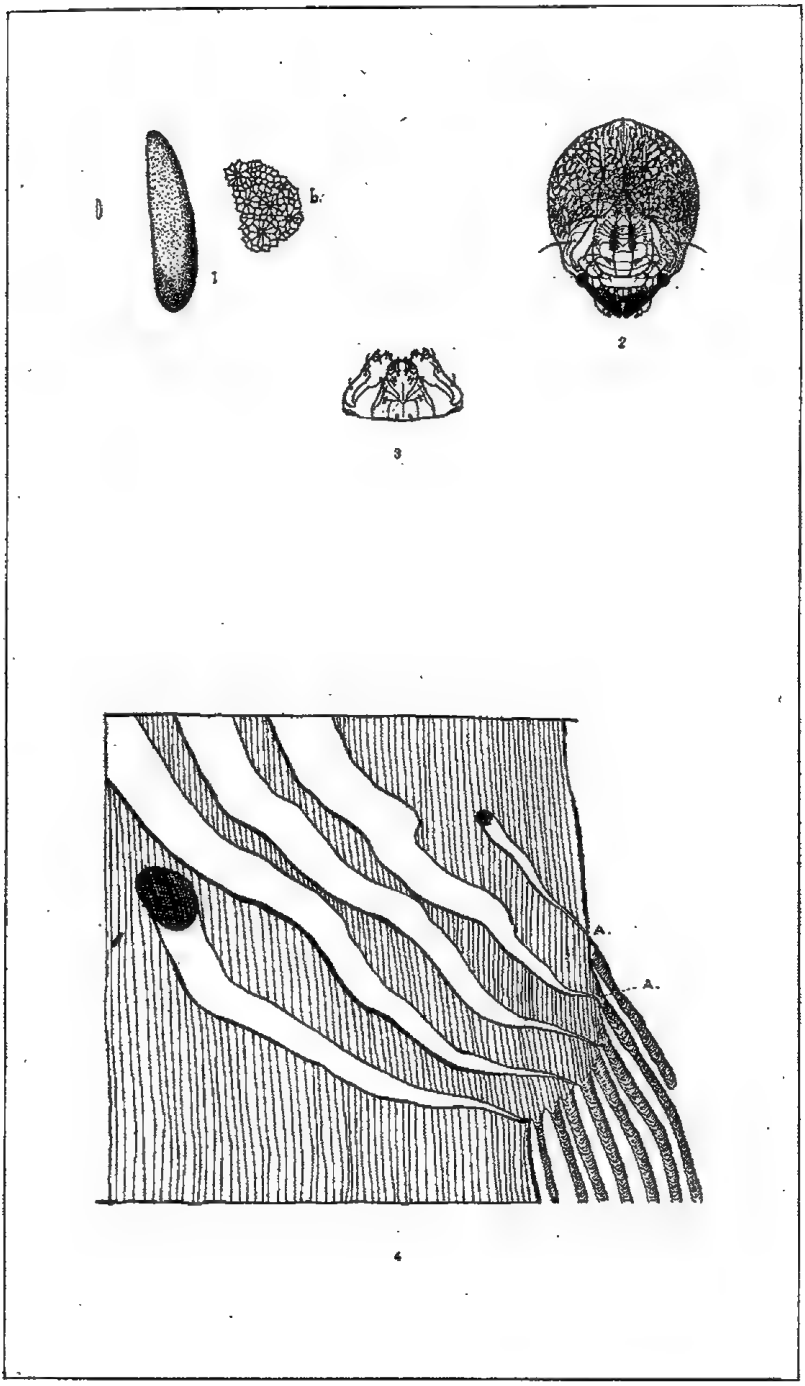


PLATE VI.

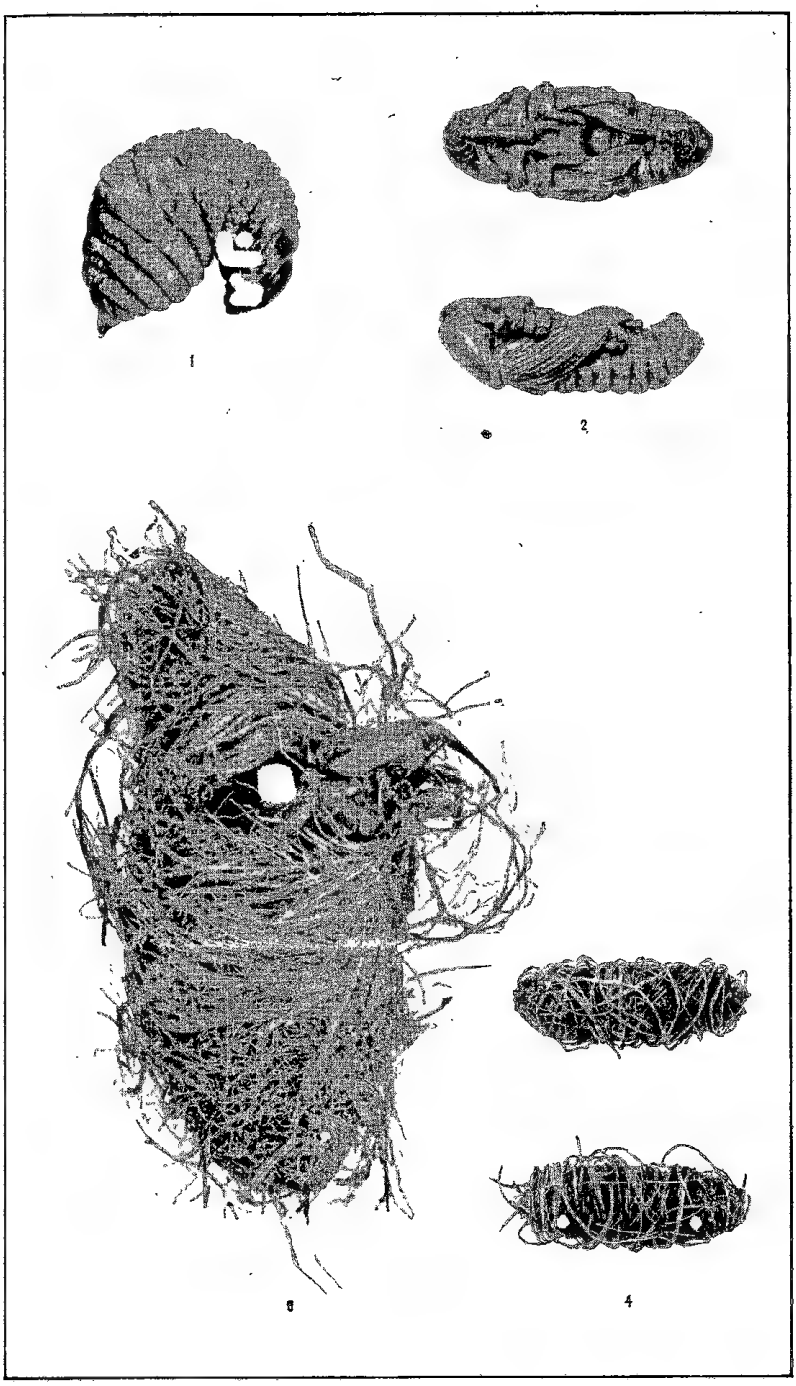


PLATE VII.

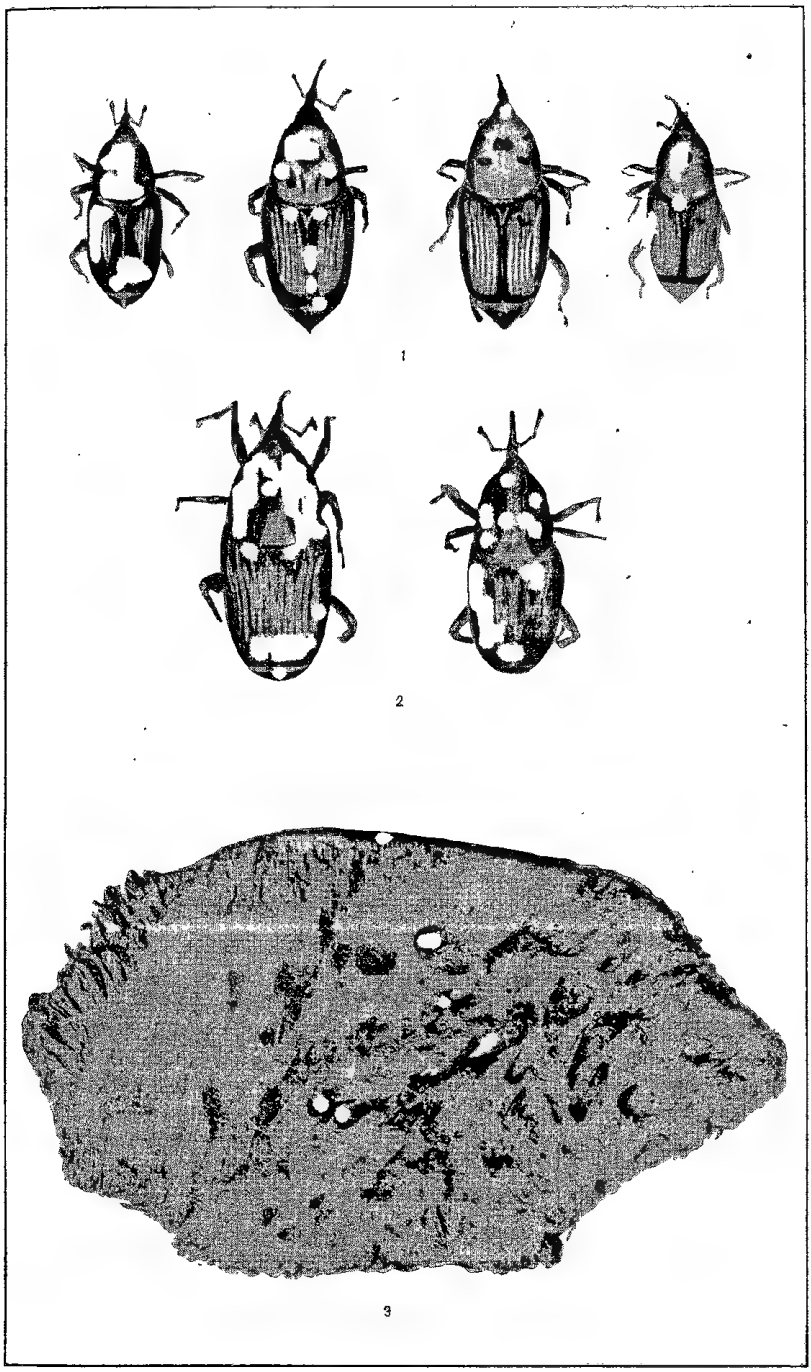


PLATE VIII.

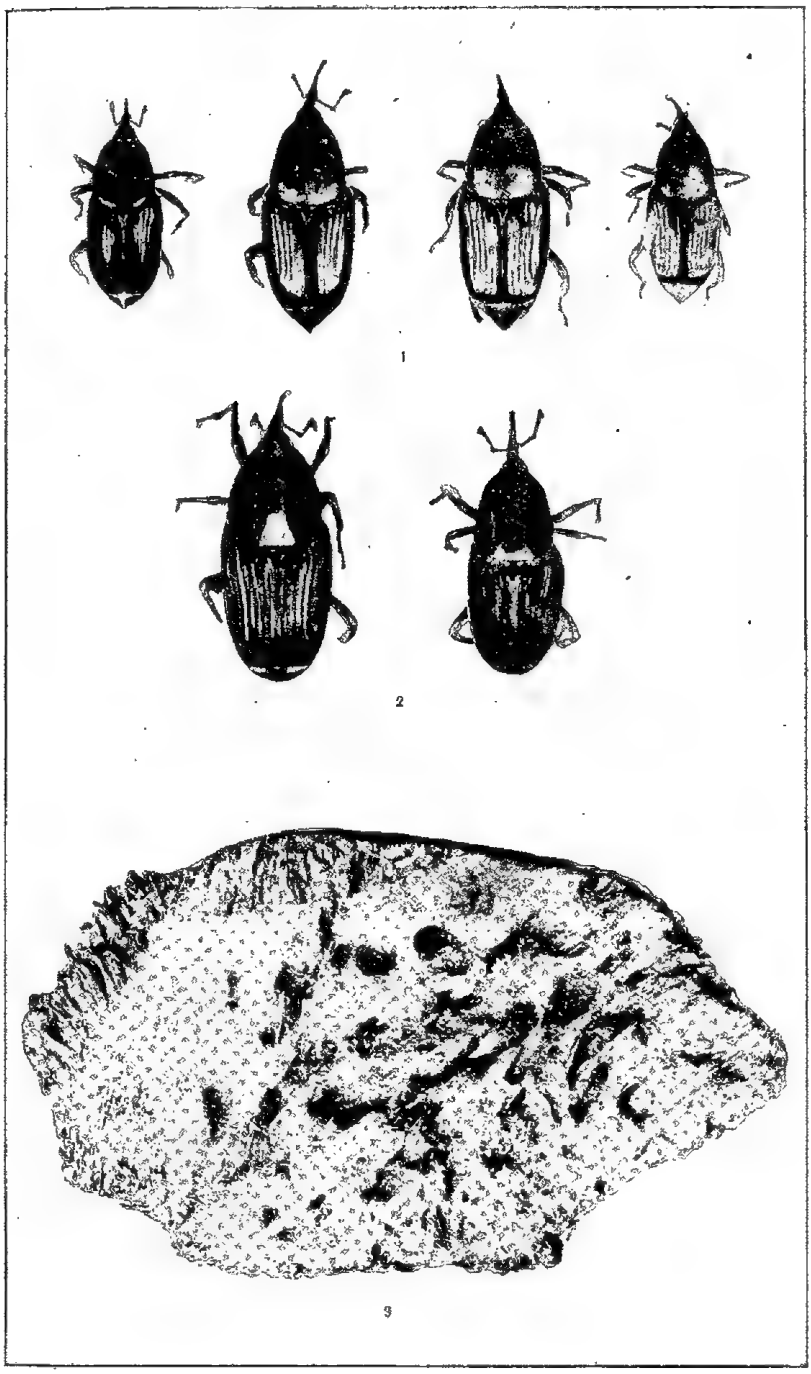


PLATE VIII.

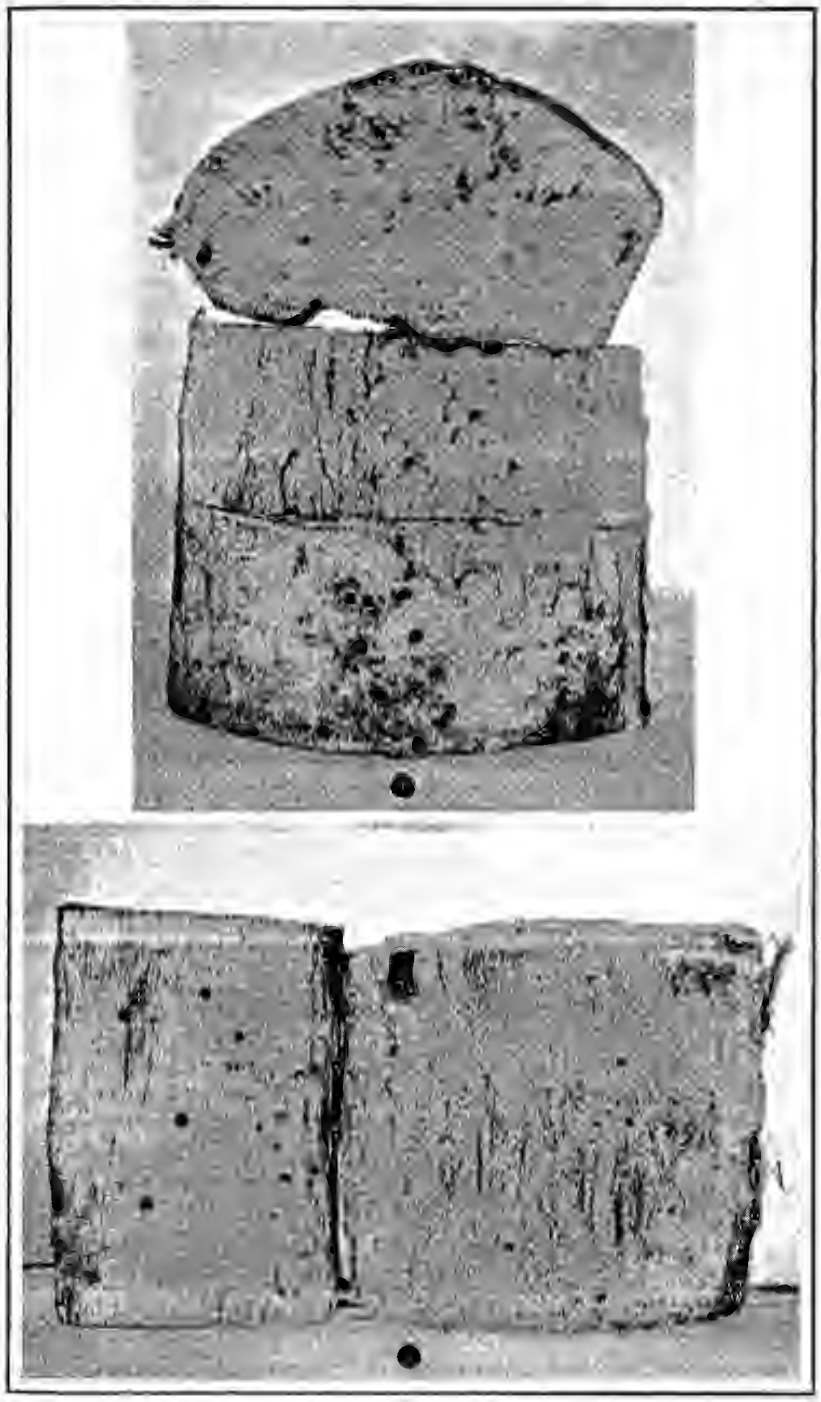


PLATE IX.

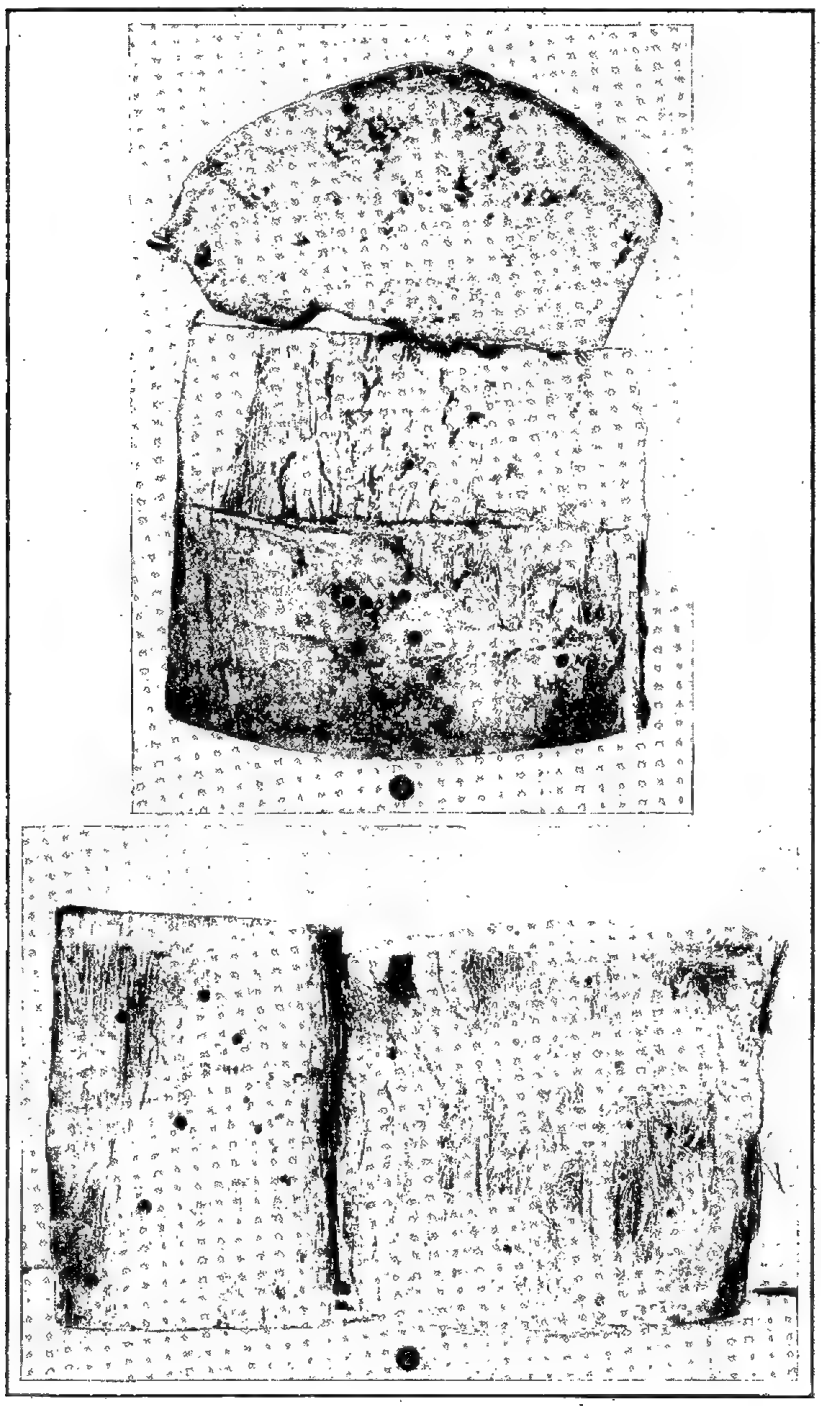


PLATE IX.

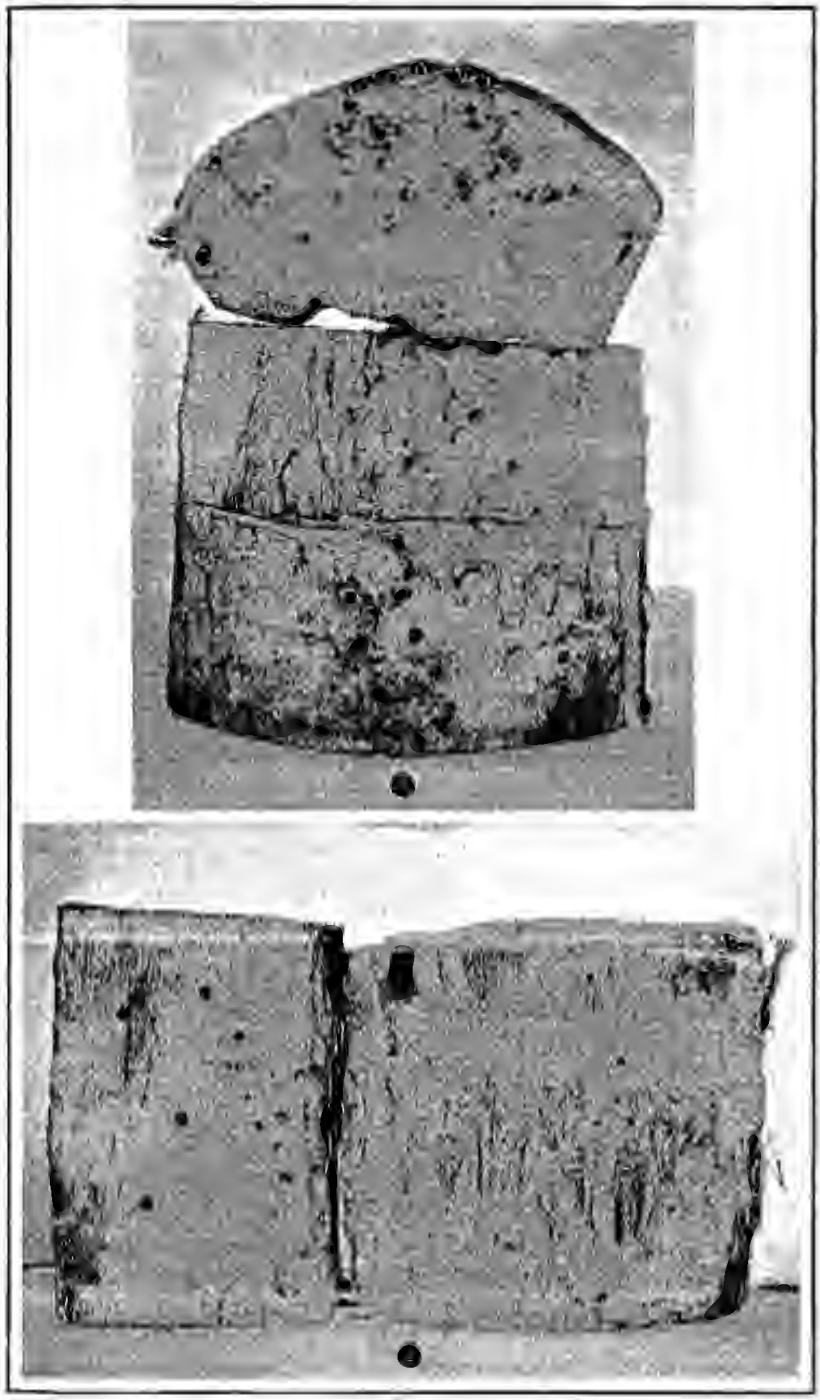


PLATE IX.

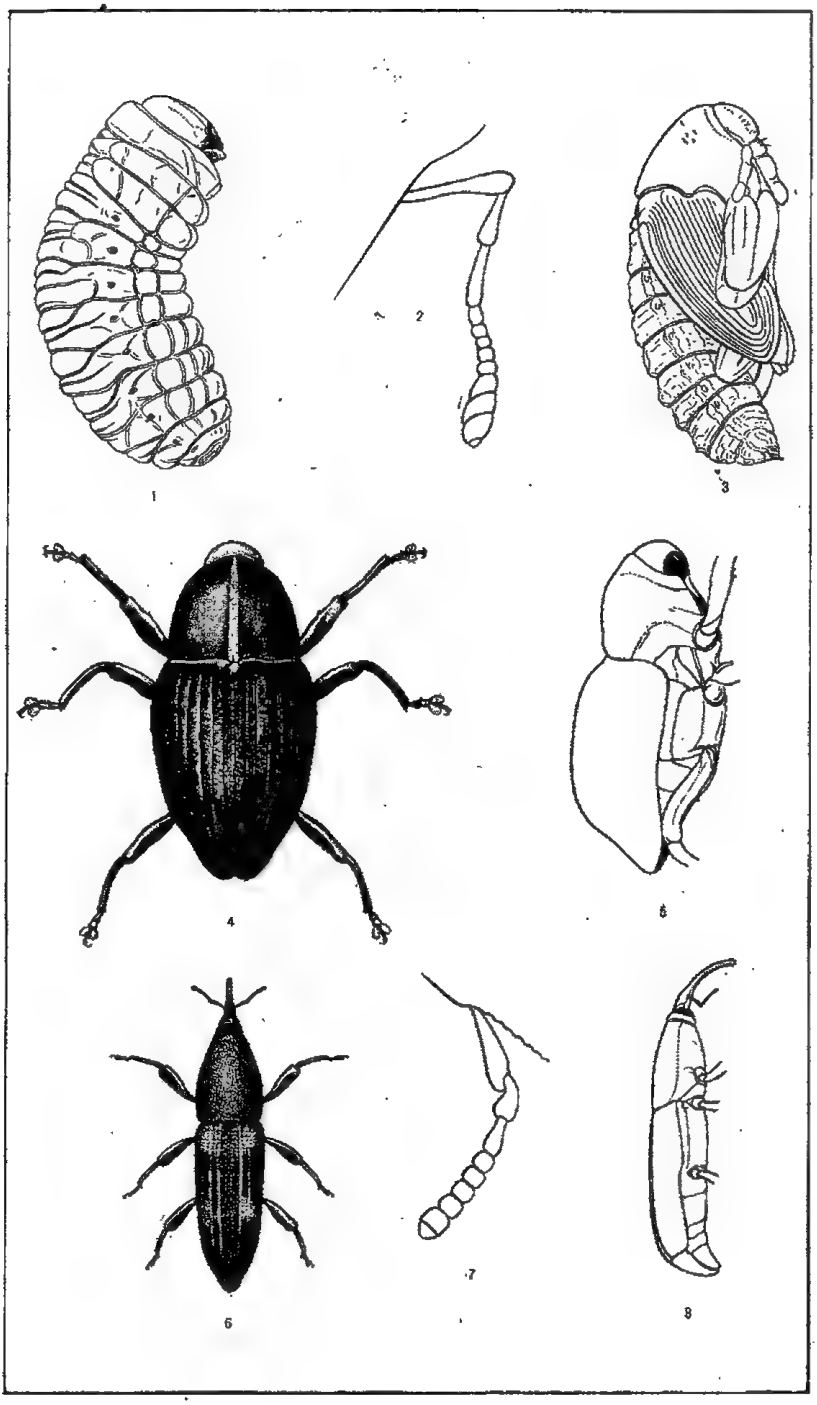


PLATE X.

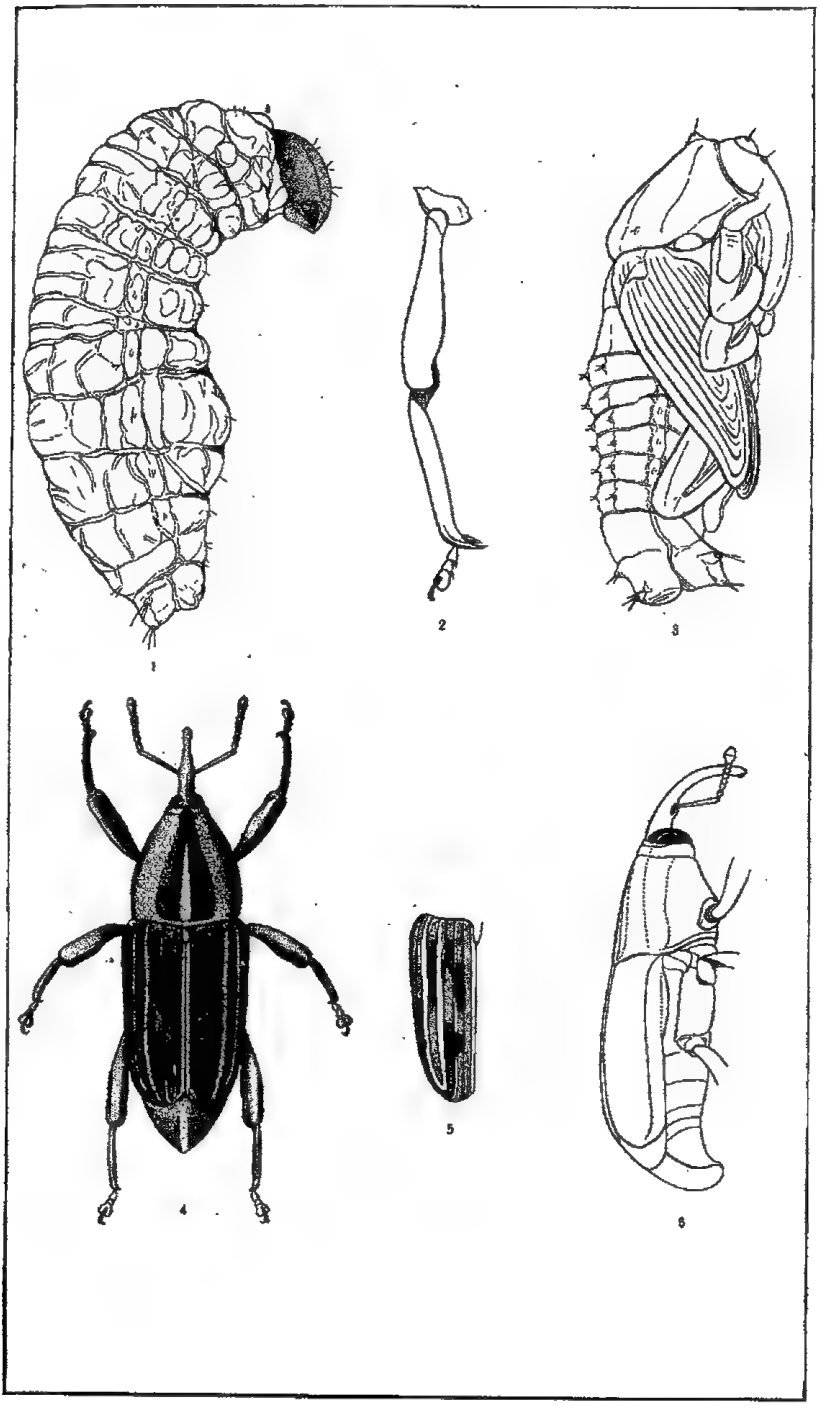


PLATE XI.

BERI-BERI IN THE JAPANESE ARMY DURING THE LATE WAR: THE KAKKE COCCUS OF OKATA-KOKUBO.

(A PRELIMINARY REPORT.)

By MAXIMILIAN HERZOG.

(*From the Biological Laboratory, Bureau of Science.*)

The affection most commonly known in European and American literature as beri-beri, and universally designated in Japan as *kakke*, is a disease generally confined to tropical and subtropical zones and particularly prevalent in certain parts of Asia, namely, Japan, China, the Malay Peninsula, the Dutch colonies, and the Philippine Islands. Especially in former years, beri-beri was exceedingly widespread in Japan, and it is from this country that most of our exact original knowledge concerning the disease has come. In this connection the work of Baelz, Scheube, M. Miura, K. Miura, and Yamagiwa, among others, should always be favorably remembered. During the late Russo-Japanese war much discussion was carried on, both in the medical and in the secular press, concerning the excellent management by the Japanese army medical corps of the sanitary affairs of the Japanese army; indeed, as it appears at present, the latter to a great extent succeeded in limiting any serious outbreak of typhoid, typhus, dysentery, or scorbutus, and in preventing entirely the appearance of cholera and plague in the rank and file of their fighting bodies. However, reports came to Manila during the second year of the war (1905) to the effect that a very large number of cases of beri-beri, occurring among soldiers returned incapacitated from the front because of this affection, were accumulating in the military reserve hospitals of Japan. As will appear from the official figures to be given below, this was indeed the case; and it was true to so great an extent that probably no outside observer, during the progress of the war, had any conception of it. It may here be stated that the total number of cases of beri-beri which developed in the Japanese army during this period is to be placed at a minimum of from 75,000 to 80,000.

However, it would certainly be doing a great injustice to blame the Japanese army medical corps for this great epidemic. There are certain points which must be considered when one is dealing with an occurrence such as this. Beri-beri is a disease the etiology of which is ill understood at the present time. We neither know its specific cause nor by what

route the specific poison, whatever it may be, enters the body. It is a well-known fact that, given certain conditions, no ordinary means of hygiene and sanitation will prevent the outbreak and the spreading of beri-beri. Such conditions are encountered in a particularly susceptible race, particularly when they are crowded together, and in certain localities. When these factors are present, nothing less than the abandonment of the locality which is apparently infected and the dispersion of the susceptible individuals will prevent the continual spreading of the disease. Of course, such measures as these could not very well be resorted to in the case of an army in the field and in the presence of the enemy. To repeat, given certain environments and certain compulsory conditions, *kakke*, in the present state of our knowledge, can not be considered to be a preventable disease.

The author was ordered to Japan to study beri-beri some time after the report concerning the presence of the disease in the military hospitals of Japan, and the information that two Japanese military surgeons claimed to have discovered its specific organism, had reached Manila. He was provided with letters of introduction from Mr. Goro Narita, Japanese consul at Manila, and he is greatly indebted to this gentleman as well as to Mr. Ishi, director of the Bureau of Commerce, Foreign Office, Tokyo, for securing permission, through the Japanese War Office, for him to pursue his studies in the military reserve hospitals at Hiroshima and Tokyo. He was also greatly assisted in the preliminary steps taken in arranging this matter by Mr. Griscom, American minister at the court of Tokyo; Mr. Huntington Wilson, first secretary of the legation, and Mr. Charles B. Harris, American consul at Nagasaki.

The author left Manila on August 15, 1905, reached Nagasaki on August 20, and arrived in Hiroshima on the 24th of the same month. At this place, situated not far from the Inland Sea, there had been a number of military reserve hospitals in operation during the war, with an aggregate capacity of 12,000 or more patients, the Japanese having transported all of their sick and wounded who could at all be moved, from the front to the home country. Hiroshima was one of the principal places for the reception of such invalids, and at that place, during the greater part of the war, there had been in operation one hospital devoted exclusively to the care of *kakke* patients. After the Japanese war office had given its permission for the pursuance of this work, the writer, through Colonel Onishi, chief medical executive officer at Hiroshima, was assigned to the *kakke* hospital to take up his studies under the direction of Surgeon Major Kokubo, who, before the outbreak of hostilities, had been professor of infectious diseases in the Military Medical School at Tokyo[1] and who was in charge of the hospital. In this institution of about 1,000 beds, there were remaining approximately 700 subacute or chronic cases of beri-beri. Connected with the hospital and situated in a separate building there was a small, though well-equipped laboratory for all ordinary bacteriologic work. Here

[1]This school was closed temporarily during the war, all of its teachers having been assigned to duty with the army.

the work in connection with the investigation of the etiology of beri-beri had been carried on by Dr. Kokubo, and here the author also worked for three weeks in isolating and examining, by Kokubo's method, the coccus which he and Okata maintained to be the specific micro-organism of beri-beri.

Kokubo had been most successful in isolating this coccus from the urine of beri-beri cases. His method was as follows: The ordinary, slightly alkaline agar medium was first melted in tubes and then poured into Petri dishes. After the agar in the plates was thoroughly hardened and dried, it was inoculated from the urine which had previously been collected in sterile receptacles, by making streaks over the surface of the media with a platinum loop. The Petri dishes were then placed in the incubator in an inverted position and examined at first after twenty-four hours and then successively from day to day. The author examined the urine from about thirty to forty cases of beri-beri and in eight was successful in obtaining a coccus which apparently was identical with the one which Kokubo believed to be the cause of the disease.

The agglutination test was regarded as the most significant feature in the identification of this coccus. Kokubo, by injecting rabbits, had prepared an antiserum which in the hanging-drop test, within one to two hours or even sooner, promptly agglutinated this particular organism in a dilution of 1–100. However, the author was not successful in obtaining this coccus from the blood of patients sick with beri-beri. He was never personally allowed to draw any blood from the sick soldiers; but Dr. Kokubo obtained some from several patients in his presence and inoculated a number of tubes with it. Dr. Kokubo's method of taking the blood was as follows:

> An area on the back, over the trapezius muscle was cleansed antiseptically and then a puncture was made with a sterile, protected blood lancet. A fold of the skin on which the small punctured wound was present was then raised between the two fingers of the cleansed hand of the operator and firm pressure applied. A considerable amount of blood could thus be obtained for the purpose of inoculating a number of tubes and procuring cover-glass preparations.

It appears that this method is not free from objection, because of the fact that it may frequently, and in a number of cases will certainly, lead to an admixture of pressed-out lymph with the blood and to the danger of contamination by means of micro-organisms inhabiting the sweat or sebaceous glands. The collection of blood by direct puncture of the median cephalic vein was suggested to Professor Kokubo, but the surgeon in charge of the *kakke* hospital refused to employ this method on his soldier patients.

The writer spent his mornings in working in the laboratory of the hospital, whereas the afternoons were devoted to bedside studies which were made while accompanying Dr. Kokubo on his daily rounds, and the latter was kind enough successively to demonstrate all of the cases at that time

present in the hospital, the patients who were more seriously and more acutely affected being repeatedly seen and examined. It was impossible to see more than one post-mortem examination; this occurred in the service of Surgeon Lieutenant-Colonel Shimada.[2] Surgeon-General Okata was also present at this autopsy and he subsequently informed the writer that from the kidneys and the cerebro-spinal fluid he had succeeded in isolating from this case the coccus which Kokubo and he had previously obtained *inter vitam* from the urine and from the blood of *kakke* cases.

In September, 1905, Okata and Kokubo published their first preliminary report on their beri-beri investigation, it appearing in the *Journal of the Military Surgical Association,* printed in Japanese. Surgeon-General Okata, who is professor of bacteriology in the Military Medical School at Tokyo, was kind enough to furnish the writer with a copy.[3] The following extract includes all of its main features:

REPORT ON THE OKATA-KOKUBO BERI-BERI COCCUS.

On examining the blood of beri-beri patients we sometimes find cocci in microscopical preparations; these are occasionally met with intracorpuscularly and sometimes outside the corpuscles. They generally appear as diplococci, but are also seen as individuals. Occasionally they are observed in the form of staphylococci. These cocci do not stain uniformly, but show an uncolored slit in the center. They are not very numerous in general, there being only one or two observed in the field. These cocci have no capsule and are not motile.

For the purpose of obtaining cultures from beri-beri cases, the blood is collected as follows: (1) The region over the trapezius is cleaned with soap and water, (2) washed with bichloride solution, (3) with physiological salt solution, (4) with distilled water, (5) with sterile alcohol, and (6) finally with sterile water. The area is then punctured with a sterile lancet and the flowing blood is utilized to inoculate a number of tubes and to make some cover-glass preparations.

The number of patients examined by this method was 129. We had 65 cases in which both microscopical cover-glass examinations and cultures gave positive results. In 34 cases both were negative, in 11 cases the microscopical examination was positive and the cultures negative, and in 19 cases the microscopical examination was negative and the cultures were positive.

Staining.—All the aniline stains, color the coccus deeply. It is well stained with Loeffler's alkaline methylene blue, and still better by the method of Semenowicz and Marzinowsky[4] (a combination of the staining solutions of Ziehl and Loeffler).

Culture.—All ordinary agar media may be used. The most suitable temperature is 30° to 37° C. When the cultures are kept at this temperature a growth, visible to the naked eye, develops after eighteen hours. At room temperature (10° C.) a very slight growth is observed after three weeks. When bouillon tubes are inoculated and placed in the incubator, a slight turbidity shows itself after fifteen hours. After twenty-four hours a grayish-white sediment forms at the bottom

[2] In the Japanese military service, autopsies can be held only after special permission from the family of the dead person is received. This permission is only very rarely given.

[3] For a translation of this report the author is indebted to Mr. C. J. Arnell, of this Bureau.

[4] *Centralblatt für Bakteriologie* (1897), 21, 874.

and some of the growth may adhere to the sides of the vessel; but the fluid is perfectly clear. The appearance presented is similar to that found in cultures of the *Erysipelas streptococcus*. When a bouillon culture is shaken, the growth forms a stringy mass of grayish-white color; and the substance which arises from the bottom of the vessel is similar in appearance to that observed in bouillon cultures of *Spirillum rubrum*. The growth is quite sticky, and on being touched with the platinum wire it forms long strings, which are not easily separated. The organism does not ferment sugar. It grows in milk without coagulating the casein.

After four days in gelatine stick cultures, kept at 18° C., there appear, along the line of the puncture, minute, whitish granules, which afterwards become larger and confluent. In three weeks the growth assumes a yellowish color. There is no liquefaction.

On agar streak cultures there first develops a grayish-white growth, which in the course of time becomes yellowish. On agar tubes kept in the incubator, after twenty-four hours there develops a granular moist growth. If examined with a magnifying glass, the colonies are seen to be granular, the margin transparent, and the center dark-yellowish.

On glycerine agar, the transparent margin is more marked and the development of the coccus is similar to that in common agar cultures. The development on urine agar is similar.

On potato, a fine, light-yellow, dry deposit develops after twenty-four hours; this does not enlarge very markedly afterwards.

On blood serum, after eighteen hours in the incubator, a very luxuriant growth develops in the line of the streak; this is shining, grayish-white in color and moist. After further time the growth spreads out from the streak in all directions in the form of branches. (In culture media to which litmus has been added, no change of color occurs.)

Urine.—The morning urine of 34 patients was collected in sterile flasks. From these specimens culture media were inoculated. In 25 of these cases we succeeded in obtaining the coccus.

Fæces.—The fæces from 44 cases were collected in sterile vessels and diluted with physiologic salt solution. We were successful in isolating the coccus on agar in 15 of these cases.

Animal experiments.—Twenty-one rabbits were inoculated; of these, 3 died. The first, inoculated with human blood, died twenty days after inoculation; the second, inoculated from the spleen of a white mouse previously inoculated from a pure culture, died nine days after inoculation; and the third, inoculated with cultures, died eight days thereafter.

Seven guinea pigs were inoculated with human blood and with the heart's blood and spleens of white mice. None of these died.

Sixty-four white mice were inoculated, of which 17 died. Thirty-one of these were inoculated with human blood, of which two died; and 33 were inoculated from cultures, of which 15 died.

Methods.—The injection into rabbits consisted of one tube of blood and sterile salt solution into the veins. In guinea pigs the same amount was injected intraperitoneally or subcutaneously. In the case of the white mice, three-tenths of a tube (0.3 cubic centimeter?) was inoculated intraperitoneally, or else a platinum loopful of the pure blood was injected subcutaneously. One platinum loopful of agar culture mixed with sterile water was injected intraperitoneally into mice, or the same amount subcutaneously. The juice from the internal organs of animals dead of the disease was also injected.[5]

[5] The last part of the report is not clear. The translation here given is as literal as possible.

(In the description of the anatomical and histological changes in the animals which died from an injection of cultures of the *Kakke coccus*, the authors do not make any statement as to the condition of the peripheral nerves.)

After having seen, and studied, more or less, all of the cases of beri-beri in the hospital in charge of Surgeon-Major Kokubo, and after having obtained a number of cultures of the coccus, which, according to Okata and Kokubo, is the cause of this disease, the author left Hiroshima about the middle of September, 1905, and proceeded to Tokyo, where, during the war, five large military hospitals had been in operation. In two of these, namely, the Shibuya and Toyama hospitals, there were still present between 700 and 800 beri-beri patients. While visiting the Shibuya Hospital the author had the advantage of having the material demonstrated by Prof. M. Miura, senior professor of pathology in the Imperial University and one of the greatest Japanese authorities on beri-beri. Professor Miura had just returned from Manchuria, where he had been ordered during the latter part of the war by the Japanese Government, and was at this time attached to the Shibuya Hospital as chief consultant for beri-beri cases. Professor Miura is still an adherent of the theory that beri-beri is a disease introduced into the human organism by food. He attributed the greater prevalence of beri-beri among the Japanese soldiers in the field during the first year, as compared with the second one of the campaign, to the following facts:

During the first year almost all of the food for the Japanese army was imported into Manchuria from Japan; during the second one, on the contrary, the commissary department (transportation department) had developed a system of purchasing and transportation in Manchuria and the surrounding parts of the continent of Asia, which enabled it to obtain its food supplies from these places without depending upon importation from Japan. Professor Miura believes that the cases of beri-beri in the field were not due to infection from those which had come from Japan to Manchuria, but to the food supply exported from Japan to the army in the field.

The author afterwards visited Toyama Hospital, where he saw a large number of beri-beri cases. This is not only the largest hospital in Tokyo but in Japan, and perhaps in the world, as it accommodates 7,000 patients. The institution is situated on a very large tract of ground on the outskirts of Tokyo, this area having formerly been occupied by the Imperial Military School for Noncommissioned Officers. It now contains from 50 to 55 different hospital and administration buildings. While in Tokyo, the laboratory of Surgeon-General Okata in the Military Medical School was also visited, and Professor Okata very kindly furnished a number of stems of his *Kakke coccus*. The following statistics as to the occurrence of *kakke* in the Japanese army during the first year of the late

war were furnished to the author during the last days of his stay by Surgeon-General Koike, chief of the medical bureau of the war office:

[Thirty-seventh year of Meiji (1904) from the beginning of the war until the month of December.]

A table showing cases of beri-beri, both those returned from the field of war and those developed at home.

Date.	Cases returned from field.		Cases developed at home.	
	New cases.	Deaths.	New cases.	Deaths.
February			30	
March	10		107	
April	66		218	2
May	106		334	1
June	258	2	313	2
July	1,602	14	424	3
August	7,960	161	596	14
September	13,505	369	373	9
October	10,811	240	326	3
November	9,344	159	296	6
December	6,682	79	320	4
Total	50,340	1,024	3,337	44

Remarks.—(1) This table after further verification will probably show a few changes.

(2) This table shows the statistics of the prevalence of beri-beri, when it was at its height last year (year 37). Although since January of the present year there has been a great diminution in the disease, exact figures can not be given, as reports from all districts have not as yet been received.

(3) Special district, regimental, and classified military reports can not yet be prepared owing to the same reasons as above given.

The *kakke* material which the writer was able to study in Japan belonged to the hydropic and to the atrophic, dry variety of beri-beri. Acute, pernicious cases were not encountered, because, of course, these had to remain at the front, most of them probably dying there. The large beri-beri material concentrated during the recent war in the military reserve hospitals of Japan, as far as the clinical histories were concerned, fully confirmed the clinical descriptions of the disease which had previously emanated from Japan. Therefore, in this preliminary report, it is not desirable to enter more fully into the subject; but the histories of three cases which were kindly furnished to us by Surgeon-Major S. Kitamura, stationed at the Shibuya Hospital, are appended. It appears that careful histories were kept of all the cases in the military hospitals, surgical as well as medical, and the whole management of these hospitals appears to have been most excellent. All of the institutions which the writer saw, excepting a very few which were older and more permanent structures, were buildings which had been erected during the

war. They were of rather cheap frame construction, but were very practical in arrangement and scrupulously clean, being well adapted to the purposes for which they were intended. Every hospital had its laboratory facilities, and in several, complete X-ray and photographic outfits were seen.

The three histories referred to follow. One of them, No. II, is illustrated by a photograph, also kindly furnished by Dr. Kitamura.

CASE NO. I.

W. S., 22 years old; infantry. The patient comes from a healthy family, has always been perfectly well, and has never suffered from any disease. About the beginning of June, while with the army in Manchuria, he noticed, without being able to assign any special reason for it, a loss of appetite, anorexia, palpitation of the heart, and precordial anxiety. After a short time, paresthesia in the tips of the fingers and in the region of the thigh was noticed, then paresis of the thigh and pain in the calves manifested themselves. The patient entered the Tokyo military hospital at Shibuya on August 6, 1905.

Status praesens: He is a man of medium size, well developed, nutrition fair. Pulse 86, full and strong. No fever. Face somewhat puffed, but no true œdema. Tongue coated; skin dry. Pupillary reaction on both sides normal. Lungs: Examination negative. *Heart:* Upper boundary of the heart's area of dullness is found at the upper margin of the fourth rib, at the left, one finger's width externally to the left mamillary line, to the right, in the midsternal line. The first mitral sound is impure. The second pulmonary sound is accentuated. There is no epigastric pulsation. Arterial sound heard indistinctly. The upper margin of the liver dullness is at the upper margin of the fourth rib. Abdomen somewhat distended; epigastrium somewhat sensitive to pressure. Appetite good. Stools, one per day, small in quantity and soft in consistency.

Disturbances of motion and sensation.—Circular hypesthetic area around the mouth. Mouth can not be very firmly closed, and it is especially the upper lip which is distinctly paretic. (Such a condition around the mouth, the report says, is very rarely seen in *kakke.*) The entire upper extremities are hypesthetic. The senses of touch, temperature, and pressure are equally disturbed. The disturbances are more marked on the anterior than on the posterior surfaces, and more on the left side than on the right one. Both flexion and extension at the elbow and wrist are diminished. Flexion of the fingers is possible, but extension is very much disturbed. All the fingers are now in a flexed position, particularly the middle and the index finger.

From the umbilicus downward anteriorly and from the gluteal region downward posteriorly to the tips of the toes, there is hypesthesia. The latter is more marked on the interior surface of the thigh than on the exterior one. The power of flexion and extension at the knee joint is diminished, more on the left side than on the right one. The dorsal flexion of the feet is decreased; the plantar flexion, however, is normal. The muscles of the calf, quadriceps femoris and adductor femoris, and the muscles of the anterior side of the forearm, are sensitive to pressure. The patellar reflex, the reflex of the tendo achilles and that of the muscles of the hand are absent, as is also that of the anterior abdominal wall, of the cremaster, and plantaris; however, the adductor reflex is preserved.

Therapy: Magnesium sulfuricum, acidum hydrochloricum.

August 13: Condition of patient improved. Pulse 72. Appetite good. Three to four stools a day. Hands and feet somewhat cyanotic. Hypesthesia around the mouth is decreased. The motility of the lower extremities is improved.

August 21: The hypesthesia around the mouth has disappeared. The mouth can be closed normally. The extension of the fingers is improved. The hypesthesia of the abdominal wall has almost entirely disappeared; that of the extremities is much improved. The motility in the knee joints is better. The feet and toes can now be moved in a normal manner. The appetite is good. Stools three times a day. The patient can now walk with the aid of a cane.

August 28: Hypesthesia is present only below the elbow and below the knee joint.

September 6: Pulse 72. The heart's dullness on the right side is found in the left sternal line and on the left side in the left mamillary line. Heart sounds pure. Second pulmonic is not accentuated. The extension of the fingers is still somewhat insufficient. The patient can now walk well with the aid of a cane; however, his gait is still a little unsteady.

CASE No. II.

S. O., 23 years old; infantry. There is no hereditary taint. The patient has always been well and has never suffered from any severe disease. Some time before the beginning of November, 1904, he noticed a loss of appetite, palpitation of the heart, and œdema of the legs. He also had a pain in the calves, and it was noticed that the patellar reflex was absent. The heart's area of dullness was increased both toward the right and toward the left. Accentuation of the heart sounds.

November 22: Pulse 108. There is no fever. There is violent palpitation of the heart. The first mitral sound is impure, and the second pulmonary sound is accentuated. There is œdema over the whole body. The abdominal wall and the lower extremities are hypesthetic.

Therapy: Infusion of digitalis and *kali aceticum*.

December 10: Appetite is good. All the symptoms are improved.

December 13: The pulse is 100 and the temperature 36.7. There is one stool daily. The patellar reflex is absent.

January 27 (1905): The heart dullness at the right side is in the mid-sternal line. This patient came to the Shibuya Hospital with the foregoing history on February 8.

Status praesens: Poorly nourished, medium-sized man. Pulse 124; temperature 36.°8. Tongue clean; extremities very much emaciated. The first mitral sound is impure; the second pulmonic sound is accentuated. Appetite good. Stools once daily. The patellar reflex is absent on both sides. Paresthesia and hypesthesia on the lower extremities are confined to the feet. The muscles of the calf are sensitive to pressure. The dorsal flexion of the left hand is insufficient. All the fingers are in a flexed position and can not be extended. However, the hand and the fingers of the right side are almost normal. The motion at the knee joints is almost normal, but the feet and toes on both sides are totally immobile.

April 10: The left boundary of the heart's area of dullness is one finger's breadth inside the left mamillary line. At the apex the first sound is dull; the second pulmonic sound is accentuated. Pulse 120. Patellar reflex slightly present. The muscles of the calves are not so sensitive to pressure as previously. The motility of the hands and fingers is improved.

Therapy: Decoctum chinæ.

April 11: The thenar and hypothenar eminences are much emaciated.

April 22: The muscles of the legs show complete E. A. R. (reaction of degeneration).

April 26: Both feet are in an equino-varus position, in consequence of which they can not be used.

May 22: The muscles of the calf are no longer tender to pressure.

May 29: The patellar reflexes have become more distinct. Flexion and extension of the knee joints are much improved; however, the feet and the toes are still entirely immobile. The right hand and fingers are almost normal in motility.

June 24: The left foot has become somewhat mobile.

July 2: The left large toe has become somewhat mobile.

August 10: The muscles of the calf on both sides are somewhat indurated. On both plantar surfaces of the feet one feels some nodular hardenings, which are quite tender to pressure, in consequence of which the patient can not walk on his soles.

August 19: Both feet and both large toes have become mobile. In attempting passive dorsal flexure of the feet and toes, one encounters strong opposition, which is caused by the tendo achilles and the aponeurosis of the plantaris. The patient complains of pain along the flexor muscles of the leg.

August 31: The motility of the toes and feet is now better and the patient can walk with the aid of crutches.

Case No. III.

T. I., 20 years old; in the military railway service. The patient comes from a healthy family and has always been well. About the beginning of August he had acute gastritis, lasting for three weeks. Shortly following it, there was noticed palpitation of the heart. The muscles of the calves were tender to pressure. There was hypesthesia of the legs. The patellar reflex had disappeared.

August 23: The patient was admitted to the hospital on this date. *Status praesens:* Medium-sized man, poorly nourished. Pulse 80, small and weak. The tongue is not coated. The voice is hoarse. The skin is dry. No œdema. The muscles and the subcutaneous tissue are somewhat diminished, especially at the extremities. *Heart:* The upper boundary of dullness is in the third intercostal space, extending to the right to the middle of the sternum and to the left to the left mamillary line. All the heart sounds are pure. The second pulmonic sound is somewhat accentuated. *Liver dullness:* The upper boundary is in the mamillary line at the upper margin of the sixth rib. The *abdomen* is moderately distended. The epigastrium and the hypogastrium are tender to pressure. A vibration can be distinctly felt at the left iliac region over the crural artery. The appetite is good. Stools twice daily. There are no dyspeptic symptoms. The hypesthetic areas are to be found only at the inner half of the feet. The right knee joint is slightly mobile; but the left one is entirely immobile. The dorsal flexion of the foot on the right side is fairly good; on the left side, however, it is disturbed. The motion in the toes is almost normal in excursion. However, the dorsal flexion of the large left toe is very insufficient. The muscles of the lower extremities are flabby and tender to pressure. This tenderness is especially marked in the adductor muscles of the thigh. The patellar reflex is completely abolished, as is also that of the tendo achilles, the abdominal wall, the cremaster and the plantaris. The triceps reflex is present.

August 30: The hypesthesia is now confined to one place, namely, the distal inner surface of the left foot. Both knee joints are somewhat immobile, particularly the left one. The dorsal flexion of the foot is improved, especially on the right side. The muscular pain is much lessened.

Therapy: Magnesium sulfuricum.

September 8: The dorsal flexion and the motility of the toes are much improved. The motility of the knee joints is now almost normal. The appetite is good. Stools once daily.

September 17: The extension at the knee joints is now almost normal in excursion.

The author, since his return from Japan, has been at work with the *Okata-Kokubo coccus* and has inoculated with it a number of monkeys and other animals. His results, as far as concerns the production in these animals of a disease similar to beri-beri in man, have not been encouraging. However, it is altogether too early to permit of any definite statements. These and other experiments will be fully dealt with in a future publication.

In concluding this preliminary report, it is my agreeable duty to express my sincerest thanks to those Japanese colleagues who have so liberally and willingly assisted me in the study of the beri-beri material to which I had access during my stay in Japan, and I wish particularly to thank Surgeons-General Koike and Okata, Colonel Onishi, Majors Kokubo, Shimada, Tanaka, Shimose, Hirai, and Kitamura, of the army, and Professors M. Miura, K. Miura, Kitasato, Shiga, and Doi, of the Imperial University and the Government Institute for Infectious Diseases, and also Captain Pershing, military attaché of the American legation, Tokyo.

The author, since his return from Japan, has been at work with the *Okata-Kokubo coccus* and has inoculated with it a number of monkeys and other animals. His results, as far as concerns the production in these animals of a disease similar to beri-beri in man, have not been encouraging. However, it is altogether too early to permit of any definite statements. These and other experiments will be fully dealt with in a future publication.

In concluding this preliminary report, it is my agreeable duty to express my sincerest thanks to those Japanese colleagues who have so liberally and willingly assisted me in the study of the beri-beri material to which I had access during my stay in Japan, and I wish particularly to thank Surgeons-General Koike and Okata, Colonel Onishi, Majors Kokubo, Shimada, Tanaka, Shimose, Hirai, and Kitamura, of the army, and Professors M. Miura, K. Miura, Kitasato, Shiga, and Doi, of the Imperial University and the Government Institute for Infectious Diseases, and also Captain Pershing, military attaché of the American legation, Tokyo.

ILLUSTRATIONS.

FIG. 1. A Japanese soldier at the Hiroshima Kakke Hospital. Atrophic, dry beri-beri; great atrophy of the muscles of the legs; contraction, with pes equinovarus. Author's original photograph.

2. Japanese soldier, Shibuya Hospital at Tokyo. Condition same as in No. 1. Photograph by Surgeon-Major Kitamura.

FIG. 1.

FIG. 2

VACCINATION AGAINST PLAGUE.

By Richard P. Strong.
(From the Biological Laboratory, Bureau of Science.)

Although the question of protective inoculation against plague has received considerable attention during the past few years and prophylactics for the disease have been recommended by Haffkine,[1] the German Plague Commission [2] (Pfeiffer and Dieudonné), Lustig and Galeotti,[3] Terni and Bandi,[4] Shiga,[5] Besredka,[6] and Gosio,[7] apparently no successful experiments have been made on human *vaccination* against the malady—i. e., protective inoculation in which the living attenuated pest bacillus has been employed.[8] It is true that in the eighteenth century some desultory attempts were made to secure immunity in man by exposing the individual to direct infection. In 1755 the Hungarian physician, Weszpremi, suggested the artificial inoculation of the pest poison in a manner similar to that which, at that time, was practiced against smallpox (*variolation*), hoping in this way to produce a mild form of the infection. In 1781, Samoilowitz, a Russian physician, inoculated himself with plague pus, suffered a mild attack of the disease, and so became immune. Therefore, he recommended that a lint compress previously saturated with the pus from a plague bubo be bound upon the arm of the person to be immunized. The skin of the individual was not to be abraded. Other observers attempted similar experiments; but many of these resulted disastrously; thus, Cerutti performed such inoculations on six persons, five of whom died of plague. Because of these results this method of immunization obviously was soon abandoned and has not since been employed. Up

[1] *British Medical Journal* (1897), part 1, 1461. Also *Lancet* (1899), 1695.

[2] *Bericht der deutschen Pest Komission; Arb. a. d. Kais Ges. Amt.* (1899), 16, 306.

[3] *Deutsche med. Woch.* (1897), 23, 227, 289.

[4] *Deutsche med. Woch.* (1900), 26, 463. Also *Rev. d. Hyg.*, Paris (1900), 22, 62.

[5] *Bericht über die Pest in Kobe und Osaka*, Tokyo (1900), 54.

[6] *Ann. Institut. Past.* (1902) 16, 918. Also *Ibid*, July (1905), 19, 479.

[7] *Ztschr. f. Hyg.* (1905), 50, 519.

[8] Kolle and Otto have called attention to the fact that the term "vaccine" is more correctly employed in the sense in which it was primarily used by Jenner and Pasteur, and should not be applied to forms of protective inoculation in which the killed organisms or their extracts are used.

to the present time no further experiments of importance with the living organism, either in a virulent or an attenuated condition, have been made on human beings, but a number of this nature have been performed on animals.

The German Plague Commission [9] (Gaffky, Pfeiffer, and Dieudonné) called attention to the fact that an ape (*Macacus radiatus*) which had been inoculated with the living pest organism and which after many days' illness had recovered, was found to be fully immune about five weeks subsequent to the first inoculation, since, at that time, it resisted the injection of an entire oese of a virulent pest culture. For the purpose of attenuating the pest bacillus, agar cultures suspended in bouillon were exposed to a temperature of 51° C. during different periods of time.

First experiment.—The culture was heated for two hours at 51° C. and afterwards proved to be sterile. A monkey, inoculated with one-fifth oese of this culture, acquired no immunity, succumbing to an injection of one oese of the virulent pest organism made twelve days later.

Second experiment.—The culture was heated for one hour at the same temperature, but in this instance all of the bacilli were not killed. A monkey inoculated with one-fifth oese of this culture also showed no subsequent immunity.

Third experiment.—The organism was heated for only one-half hour at 51° C. In this instance many of the bacilli were also not killed, and the monkey inoculated with one-fifth oese of this culture died of pest four days after this primary injection.

In the foregoing observations, as was demonstrated by further experiments, the pest bacillus was not attenuated, but was either entirely killed or had retained its full virulence. In the second experiment, while the living organisms were still present in the vaccine, their number was obviously too small to produce the desired immunity or even to give rise to any decided reaction in the animal. This was conclusively shown to be the case by the following observation, in which a two-day agar culture was suspended in 5 cubic centimeters of bouillon and then further diluted with saline solution, so that ape A received 1 cubic centimeter in a dilution of 1 to 100,000; ape B, 1 cubic centimeter in a dilution of 1 to 10,000, and ape C, 1 cubic centimeter in one of 1 to 1,000, subcutaneously. No apparent reaction occurred from these injections and only ape C showed any traces of immunity, this animal remaining alive for nine days following the injection of 1 oese of the virulent pest bacillus (although it succumbed after this period), while the other two animals died on the third day after such an inoculation.

In another series of experiments, the commission attempted to obtain an attenuation of the organism by its exposure to the action of carbolic acid. However, no loss of virulence resulted by this method, the animals injected with the cultures which had been so treated all succumbing to pest infection. On account of the difficulties encountered in the attenuation of the plague bacillus or in obtaining cultures already attenuated,

[9] *Loc. cit.*, 303.

the commission abandoned the idea of employing the living organism for the purpose of obtaining immunity against the disease.

Albrecht and Gohn[10] (the Austrian Plague Commission) performed experiments with attenuated plague cultures on eight guinea pigs and twenty-seven rats. Of the guinea pigs inoculated, either subcutaneously or intraperitoneally, with the living organism of reduced virulence, and later repeatedly reinoculated with increasing doses of a virulent culture, five finally died and three remained alive. The immunity was found still to be present in some of the animals seven months after the vaccination. It is not altogether clear why repeated and increasing amounts of the more virulent organism were injected in testing the immunity of the guinea pigs, unless the authors felt uncertain of the exact virulence of the culture which they used or of the method they employed.

Kolle has recently criticised some of this experimental work more in detail, referring particularly to the fact that the animals were, at least in some cases, reinoculated with the virulent culture at too short a time after the vaccination, for the results of the experiments to be conclusive, since in some instances they might have been suffering from chronic pest at the time of the reinoculation. Besides this, the quantity of the bacteria injected in testing the immunity of some of the animals was very large. Twenty-one of the twenty-seven rats which Albrecht and Gohn vaccinated remained alive after reinoculation with the virulent organism. Only one experiment was performed upon a monkey; in this instance the animal was apparently immunized successfully, but it finally died of tuberculosis. Albrecht and Gohn conclude that immunity can be obtained in animals by employing the living pest bacillus, but that this process must be carried on in a careful manner in order obtain a fair degree of protection. From their experiments they were unable to decide whether the immunity caused by the injection of the killed pest bacillus was fully as great as that which resulted from the inoculation of the living organisms.

Yersin and Carré[11] also performed experiments upon the immunization of rats with attenuated strains of the plague bacillus. They finally obtained a culture of such diminished virulence that only one-fifth of the animals vaccinated with this organism succumbed to the effects of the injection. A series of twenty-five rats was inoculated with this culture. Three of these died from the effects of the vaccination and about three weeks later the remaining twenty-two animals were inoculated with the virulent pest organism, after which only one succumbed. A second series of twenty rats was inoculated with the same bacillus. Ten of these, which survived the vaccination, were later inoculated with the virulent

[10] *Denkschrift d. math.-naturw. Klasse d. Kaiserl. Akad. Wien* (1898 and 1900), **66**, 807.

[11] *Congrès International de Médecine, Section de Médecine et Chirugie Militaires. Sous-section Coloniale*, Paris (1904), 54.

pest culture, when all proved to be immune. Only two control animals were inoculated in each of these series. This number was hardly sufficient to render the experiments conclusive. The authors do not state the size of the dose, the virulence of the organism used in testing the immunity, and the method of inoculation. When this same attenuated bacillus was allowed to grow continuously on artificial media during forty to fifty days it was claimed that its virulence was greatly decreased, since of thirty rats inoculated with such a culture, none died. Later, on testing fourteen of these animals with the virulent pest organism, five succumbed. Seven apes were inoculated with another plague culture which, when injected into rats, killed from 40 to 50 per cent of these animals. None of the apes died from the effects of the vaccination. Apparently only two of these animals were subsequently tested for their immunity; these developed localized buboes, but recovered. Since the single ape used for control purposes also did not die we can draw almost no conclusions from these experiments. The amount of the organism employed in the vaccination or in the testing of the immunity is not stated. Yersin apparently was inoculated with the most attenuated culture (fifteen days old), but the size of the dose used in the vaccination is not given. Only very slight symptoms developed. At the same time, ten rats were injected with this culture but none died. The immunity of these animals had not been tested at the time the paper of these authors appeared. Although these experiments were reported in 1900, I have been unable to find any further reference made to them since that date either by Yersin or his colleagues.

No extensive or convincing experiments in regard to the value of the employment of the living attenuated cultures in the immunization even of animals against plague had apparently been undertaken until Kolle investigated this subject.

In 1902 and 1903 Kolle and Otto [12] inoculated eighteen guinea pigs subcutaneously with an attenuated culture of the pest bacillus. The organism was an old laboratory culture in which the reduction of the virulence had, in some unknown manner, taken place during its growth on artificial media. Buboes, which later discharged and healed and in the pus from which a few bacilli were present, developed in the animals, but they showed no other evidence of sickness and subsequently entirely recovered. The animals were reinoculated two, three, and eight months later with one-twentieth to one-fiftieth oese of a pest culture (of which one one-hundredth oese represented the fatal dose for a normal guinea pig). Seven of the animals remained alive.

In a large series of rats immunized by various methods, the loss from inoculation, with the living attenuated cultures, was 2.3 per cent; with the killed agar cultures, 33.3 per cent; with Haffkine's prophylactic, 38.5

[12] *Ztschr. f. Hyg.* (1903), **45**, 512.

per cent; and with Lustig's prophylactic, 12 per cent. After reinoculation (for the purpose of testing the immunity of the rats), 45 per cent of those inoculated with the attenuated living organism, 21.9 per cent of those which had received the killed agar cultures, 22.2 per cent of those injected with Haffkine's prophylactic, and only 12 per cent of those which were given Lustig's preparation remained alive.

However, Kolle and Otto's further experiments on guinea pigs are more interesting and conclusive; for, as these authors have remarked, a preparation which is recommended and used for the control of plague in human beings should develop a pronounced protective effect on the guinea pig, the animal which is the most susceptible to plague. Among fifty-nine guinea pigs which they immunized with other attenuated living cultures, thirteen died and two were killed for control purposes. The remaining forty-four (75 per cent) were tested for their immunity. All of these had received but a single injection of the weakened culture; however, upon their reinoculation three, four, and eight months after this vaccination, twenty-eight (63.6 per cent) remained alive after the injection of the virulent organism. Six other guinea pigs received somewhat larger doses of a less attenuated culture, and at the same time an injection of serum; five survived the vaccination and one died. The former, upon reinoculation with the virulent organism, later all proved to be immune. Including the loss during vaccination, there finally remained alive, after testing the immunity, 50.8 per cent of the whole series.

Attempts were made to immunize twenty-six guinea pigs with *killed* agar cultures, amounts as large as from one-half to one entire agar culture being injected subcutaneously. During the process of immunization, four of the animals died; of the remaining twenty-two, only two (7.7 per cent of the whole) proved to be immune on subsequent testing. Hardly more favorable results were obtained in the experiments in which *killed bouillon cultures* were employed. Twenty animals were inoculated with Haffkine's prophylatic; two of these died during immunization and of the remaining eighteen, only two (10 per cent) remained alive after reinoculation with the virulent organism. The method of immunization, in which Haffkine's prophylactic was first injected and later followed by the introduction of the attenuated living cultures, did not give as favorable results as did the living avirulent organism alone.

In December of the past year (1904) Kolle and Otto[18] in further detail reported upon the immunization of guinea pigs with the attenuated pest bacillus. Among thirty-four of these animals immunized with such a culture (*Maassen V*), of which none died during the process of immunization, twenty-one were reinoculated with a virulent organism one to four months after their vaccination, and of this number sixteen (76 per cent) remained alive, and five died. Nine other guinea pigs were inoculated with the avirulent culture, and at the same time with plague

[18] *Ztschr. f. Hyg.*, December (1904), **48**, 399.

immune serum; all proved to be immune upon reinoculation with the virulent pest bacillus. The organism with which these vaccination experiments were performed possessed so little virulence that from two to three living, agar slant cultures, when injected into a guinea pig of 250 grams' weight, did not cause the death of the animal. In the experiments of Kolle and Otto on the vaccination of monkeys, almost all of the animals died from the effect of the attenuated culture or succumbed to intercurrent disease.

Other experiments on guinea pigs, in which repeated inoculations of the *killed* cultures of the plague organism were employed, were also performed. The animals were first injected with one, then with 1.5, and finally with two killed agar cultures, or with 1, 1.5, and 3 cubic centimeters of Haffkine's prophylactic. In the process of immunizing twenty guinea pigs, six of the animals died from the effects of such large doses of the killed bacteria. The immunity of the remaining fourteen, six weeks after the last injection, was tested with the living virulent plague bacillus, when only one animal remained alive and proved to be immune.

Therefore, Kolle emphasizes the fact that, if such large and repeated doses of the killed pest organism fail to immunize such small animals as guinea pigs, it seems unreasonable, from such a method, to expect very favorable results in man, particularly since in the latter case the amount of the bacteria inoculated is so much smaller in proportion to the body weight. It is to be noted that in immunizing the guinea pigs similar or larger doses of killed organism were employed than have been recommended for protective inoculation in human beings.

It is not my purpose here further to enter into the discussion of the protective value of the different prophylactics recommended for the immunization of man against plague. I had already concluded from animal experiments, as well as from the fact that a number of persons who had received several injections of Haffkine's prophylactic later sickened and died with plague that *the killed pest organism* constituted for man a far from satisfactory protective against this disease. On the other hand, the experiments which Kolle and his pupils had performed on guinea pigs seemed so conclusive in regard to the value of *the living attenuated cultures* in the immunization of these animals that I felt convinced that, if these cultures were of so low a virulence, or could be further so attenuated as to warrant their use in man, a higher degree of immunity could almost certainly be obtained with them than by the employment of the *killed* bouillon or agar cultures.

Accordingly, when Professor Kolle, after some correspondence on the subject, kindly offered me cultures of these attenuated pest organisms which had been employed in his experiments on guinea pigs, for use in the vaccination of human beings, I decided to carry on this work. My experiments in vaccination in man and animals have been performed with three attenuated strains of the pest bacillus, *Maassen Alt* and *Maassen V*

of Kolle and an old Manila culture which had been grown continuously upon artificial media for three years and whose virulence has been still further reduced by artificial means. The attenuation of this last culture was further brought about by growing the bacillus at a temperature of from 41° to 43° C. in flasks of alcoholic bouillon for three weeks at a time, as recommended by Otto. Cultures from these flasks were then inoculated on agar for many generations, a fresh generation being made every day for several weeks, and the organisms always cultivated at the same high temperature. Beginning with 0.05 cubic centimeter of absolute alcohol and 50 cubic centimeters of bouillon, the amount was gradually increased in successive cultures up to 5 cubic centimeters in 50 of bouillon. Before making inoculations in man, the action of the attenuated culture was of course carefully tested in animals.

In the present paper it is merely my desire to call attention to the fact that vaccination in man can with safety be performed with attenuated cultures of the living plague organism, and therefore only the human inoculations undertaken with one strain of this bacillus will be referred to.[14] The organism in question (*Ma. V*) possesses so little virulence that in a series of twelve guinea pigs and thirty monkeys inoculated with from one to two entire agar slant cultures, not one succumbed from the effects of the inoculation.[15] It was with this culture that the first experiments were performed in human beings. Since I believed that the guinea pig is an equally if not even a more susceptible organism than man to the pathogenic action of the plague bacillus, it was presumed that if this animal could invariably withstand the action of such large amounts as two whole agar slant cultures of the organism, much smaller quantities could be inoculated into human beings with safety, and indeed, before performing the experiments on man, I felt thoroughly convinced of this fact; nevertheless, the human inoculations were performed as carefully and with as much deliberation as possible.

The first injections were carried on upon prisoners under sentence of death; in the first case one-hundredth oese of the attenuated culture was inoculated subcutaneously without any noticeable effect. After ten days, ten other individuals were inoculated with the same dose, in order to demonstrate that no special natural immunity against the plague organism had been existent in the first instance. In this manner the amount of living organisms given was gradually increased, a single person being first inoculated with the larger dose and then, after it had been observed that no unfavorable effects occurred, from five to ten other

[14] The results of these experiments were communicated in a paper read before the Manila Medical Society at its meeting on November 6, 1905.

[15] But one of the animals of the series perished. Monkey No. 1299 died about twelve hours after inoculation with the avirulent pest organism, of a staphylococcus and streptococcus pyemia which had existed prior to the inoculation. A large suppurating wound existed over the abdomen.

persons were also treated with the same amount of the vaccine. This method of procedure was adopted in order to minimize the danger of inoculating a very susceptible individual with a dose which might prove disastrous. It was argued that if ten persons selected at random withstood the inoculation of a certain amount of the organism without developing unfavorable symptoms, a single individual, also selected at random, could probably receive a slightly larger dose without great danger. In this manner as mentioned the dose was gradually increased until one whole agar slant was inoculated. No attempt has been made to inject a larger amount of the organism, since from experiments performed on animals it has been concluded that a sufficient immunity in man will probably result from an inoculation of this quantity. Up to the present time forty-two persons have been injected with this large dose (one twenty-four hour agar slant culture) of the living bacillus, and, although the inoculations which I include in this report were all performed more than two months ago and the individuals treated have been under constant surveillance, I have no accident to report.

Surprising as it may seem, the injection of these large amounts of the living plague organism have not given rise to any very severe reactions. A few hours after the inoculation, the temperature of the individual usually begins to rise. When the injection has been given in the morning the fever may, on the evening of the first day, reach 38.9° to 39.4° C. (102° to 103° F.), but rarely has it touched 40° C. (104° F.). On the following day, in none of the cases was the temperature above 38.9° C. (102° F.) and usually not above 37.8° C. or 38.3° C. (100° or 101° F.) and on the third one it generally was normal. Occasionally the cases showed a moderate leucocytosis after the large injections. The organisms were always suspended in 1 cubic centimeter of .085 saline solution and the inoculations were made deeply into the deltoid muscle. On the day after the vaccination there usually was distinct induration about the point of injection, with some soreness on pressure, but these symptoms subsided in one or two days. No suppuration ever occurred. A careful study of the blood serum has been made in twenty-nine of the human cases; agglutintative tests have been performed with the virulent plague organism and the anti-infectious power of the serum has been tested in rats. A detailed report of all the experimental work will appear in a future number of this JOURNAL.

It was interesting to observe the length of time during which these avirulent pest organisms remained alive in monkeys after subcutaneous inoculation, and for this purpose a series of ten animals was injected upon different days, the cultures being taken at periods of from one to twenty-four hours after the inoculation. The abdomen of the animal was first shaved, and the injection made subcutaneously. The skin was then carefully massaged until apparently the fluid was completely absorbed. At the time the culture was to be taken, the skin of the abdomen

was scrubbed several times with ether and alcohol and a small incision made with a sterile knife through the dermis. The cultures were then made from the drops of blood which oozed from the incised wound. Usually, when the injection is made beneath the skin of the abdomen, after a few hours an œdematous swelling, which may not entirely disappear for from twelve to twenty-four hours, appears near the point of inoculation.

The different series of cultures made from the animals have shown that six to eight hours from the time of inoculation the organisms are still very numerous in the tissues, after which time they gradually disappear, so that the cultures made twenty-four hours subsequent to the injections remain sterile. It seems probable that the more resistent organisms are those which remain alive the longest, and that there is here a true survival of the fittest. A trial is therefore being made to ascertain whether it is possible to increase the virulence of these attenuated pest strains by such a procedure. As soon as the cultures made from the blood of one animal develop, they are inoculated into another monkey, and so on through a long series. It seems possible that such a method may have certain advantages over that in which the organisms are inclosed in celloidin sacs and placed in the abdominal cavity of animals. Kolle and Otto have shown that it is very difficult or impossible to increase the pathogenesis of more virulent strains of pest bacilli [16] by repeated passages through guinea pigs; but this need not necessarily be true with more avirulent strains.[17]

It may be questioned whether the organism which has been employed in these human inoculations is really a strain of *Bacillus pestis*. Therefore, while it is not considered necessary in detail here to relate the immunity reactions, morphology, etc., of this organism, it may be stated that unquestioned proof of this lies in the fact that I have vaccinated both guinea pigs and large numbers of monkeys with this culture and have later shown them to possess high and undoubted pest immunity by subsequently inoculating them with large amounts of virulent plague bacilli. Indeed, with no other method of inoculation have I been able to obtain such favorable results as with this pest *vaccine*.

Allusion has been made in other places to the similarity between certain of the immunity reactions of plague and of rinderpest. The present method of human vaccination against plague may be compared with that first recommended by Robert Koch in the immunization of cattle against rinderpest. Koch used the bile of animals which had died of this disease and in which *perhaps* the living attenuated organism of rinderpest exists. Doubtless a higher immunity in man against plague could be obtained were a more virulent culture of the pest bacillus

[16] Those which kill the animal.

[17] Those which are not capable of causing the death of the animal, even in large amounts.

injected and at the same time a dose of antiplague serum inoculated,[18] but there undoubtedly would be a mortality by such a method, just as in rinderpest there is always a certain fatality among cattle when the virulent blood and antirinderpestic serum are injected simultaneously, and therefore such a method of immunization, though valuable for animals, can not be recommended for man. Moreover, it is questionable whether any higher immunity in human beings than that which can be obtained from the attenuated, harmless culture is usually necessary.

In publishing for the first time the results of my vaccinations in human beings against plague, I wish to sound a note of warning against the employment of other strains of living pest bacilli for this purpose. Inoculations should not be made in man, unless the investigator can guarantee the organism with which he is working to be of sufficient attenuation to be no longer dangerous for human beings. Strains of the bacillus which invariably no longer kill guinea pigs in doses of two entire forty-eight hour agar slant cultures are probably safe in very small amounts for human beings. Unless excessive precautions are taken in inoculations of living plague bacilli, disastrous results will surely follow. It seems probable that by pursuing proper methods for sufficient periods of time all strains of pest bacilli perhaps can be attenuated sufficiently to be safe for the purposes of human inoculation; although in some instances it may take several years to bring about such a result. I believe that the avirulent pest culture with which these experiments were performed is at present hardly more dangerous for inoculation in man than is attenuated vaccine taken from human beings suffering with smallpox.

In concluding this preliminary report I wish to express my thanks to Professor Kolle for two of the avirulent pest cultures and again to call attention to the fact that it was the careful and extensive work on this subject which has come from his laboratory during the past two years which convinced me of the value of the living attenuated plague cultures in immunization and which caused me to undertake this further study of the question in man and animals.[19]

[18] My experiments have convinced me that in pest (as in cholera) unquestionably a higher immunity in animals can be obtained with the more virulent organism than with the less virulent one. The pest bacillus therefore may differ from the typhoid organism in this respect, since Wasserman has shown that this phenomenon does not necessarily result with the latter bacterium.

[19] While this article was in press there reached Manila the *Centralblatt für Bakteriologie* [(1905), **39**, 610] containing a report of the experiments of Hueppe and Kikuchi on immunization of animals against plague by means of "aggressin" obtained according to the method of Bail [*Archiv. f. Hyg.* (1905), **52**, 272]. The value of immunization with plague "aggressin" prepared after the method of Wassermann [Wassermann, and Citron, *Deutsche Medizinische Wochenschrift* (1905), **31**, 1101] as compared with that of the living attenuated plague organism will be discussed in the more complete report.

PHILIPPINE WOOD OILS.

By A. M. Clover.
(From the Chemical Laboratory, Bureau of Science.)

In a study of tropical forest products there are encountered as exudations from trees a great variety of substances, all of which may appropriately be termed resins. They appear and are collected in different physical conditions which are modified by the rapidity with which they issue from the tree and the rate at which they dry or harden thereafter. The latter function is dependent upon the relative amounts of water, oil, and solids which are found in the resins and upon the chemical composition of the oil. Accordingly, many resins are encountered in the solid form and contain very little volatile matter, whereas others collect upon the tree in a plastic condition and still others harden so slowly that they are removed as fluids.

The members of the latter class which have been differentiated in a study of the resinous products of the Philippine Islands have shown among themselves a similarity in chemical composition and a likeness to other known products to such an extent that they may appropriately be placed in a class by themselves and designated as *wood oils* (a term sometimes applied to gurjun balsam). A *wood oil* is therefore a fluid resin of very slight "drying" power and containing a high percentage of volatile matter; the oily portion of this volatile matter sometimes is as much as 75 per cent of the total resin and consists entirely of bodies belonging to the sesquiterpene group. In no case has a low-boiling or terpene oil been observed in this class of products; but on the other hand the viscous resins nearly always contain terpenes and the relative amount of oil found in them seldom exceeds 25 per cent.

OIL OF SUPA.

The tree (*Sindora wallichii* Benth) yielding this oil is said to be widely distributed over the Islands. The sample examined was sent to the laboratory from the Province of Tayabas, where in certain localities it is reported to be used as an illuminant, but no information concerning its use in other parts of the Islands could be obtained. A freshly cut tree, so it is stated, yields about 10 liters of the product, to obtain which it is

necessary to make a cavity in the trunk. Botanical specimens of the tree from which the sample under consideration was taken accompanied it and they were identified by Mr. Merrill, of this Bureau, it being the species given above. The oil is quite mobile, perfectly homogeneous, light yellow in color, with a slight fluorescence and a feeble but characteristic odor.

Specific gravity, $\frac{30°}{30°} = 0.9202$. Optical rotation, —31°.3 (10 centimeters, 30°). When cooled below 20° it begins to deposit white, flaky crystals; these increase in amount as the temperature is lowered. The crystals are those of a hydrocarbon, melting at 63° to 64°, and occurring in the oil to the extent of a few per cent. The oil completely dissolves in all the ordinary solvents, excepting alcohol, which causes the separation of the white crystalline hydrocarbon already referred to. Oil of Supa takes up oxygen slowly from the air and finally hardens, several weeks being necessary for the drying of a thin film.

Volatile portion.—When Oil of Supa was subjected to steam distillation, a colorless oil was slowly carried over, the process being continued until most of the latter products had been removed. On allowing the distillate to stand until it was perfectly clear it showed a rotation of —21° (10 centimeters, 30°). It was impossible to obtain a constant-boiling product from the latter upon fractioning under diminished pressure. At 40 millimeters nearly all of the oil passed over between 143° and 149°, the residue apparently being as fluid as the distillate and indicating no polymerization. The distillate had a specific gravity of $\left(\frac{30°}{30°}\right)$ 0.9053 and when redistilled it boiled between 255° and 267° (760 millimeters) with practically no residue. In order to save time it was found to be more convenient to remove the volatile oil by direct distillation under reduced pressure. With a pressure of 40 millimeters the temperature of the distillate gradually rose to 170°, at which point nearly all of the volatile portion was removed, the total quantity in the receiver consisting of about 73 per cent of the original sample, and containing only a slight amount of water. The residue was light brown in color and became semisolid on cooling; it dissolved in all the ordinary solvents, excepting alcohol, which separated the solid hydrocarbon referred to above. The distillate was colorless and practically remained so on standing in a closed vessel. It had the feeble, but characteristic, odor possessed by the steam-distilled oil, and on refractioning at 40 millimeters it almost completely passed over within 7°. No low-boiling substance was noted, and after repeated distillation it was not found possible to obtain a much more constant-boiling product. The oil probably is a mixture of sesquiterpenes, as cadinene was proven to be present. The absence of bodies of an alcoholic nature was demonstrated by the fact that neither sodium nor phosphorus pentoxide in benzol have any action on the oil. On passing hydrochloric acid gas into a solution of the distillate in acetic

acid, cadinene hydrochloride, as described by Wallach[1] (melting point 117° to 118°), separates.

0.1878 gram substance gave 0.1920 gram AgCl.

Required for $C_{15}H_{24}$, 2 HCl		Found
	Per cent.	Per cent.
Cl.	25.6	25.3

The hydrobromide was also prepared and recrystallized from ligroin. Its melting point (118° to 125°) corresponds to that obtained by Wallach.

On saturating a solution of one part of the distillate in four parts of glacial acetic acid with hydrochloric acid gas at 5°, the hydrochloride soon separated in considerable quantity. The solution was kept saturated for several hours and then allowed to stand for a day. It was then cooled, filtered quickly with suction, and washed with alcohol. Twenty grams of the hydrochloride were obtained from 45 grams of the oil. To convert the hydrochloride into cadinene it was heated with excess of aniline at 150°; aniline and aniline hydrochloride were removed with dilute acid and the remaining product was distilled with steam and redistilled under diminished pressure. It boiled at 164° to 165° (38 millimeters). Not enough of the substance was at hand for a determination of its specific gravity. Rotation, —39° (5 centimeters, 30°). It will be noted that the treatment with hydrochloric acid gas only partially converted the oil into cadinene hydrochloride, but a separate determination of the total amount of hydrochloric acid which the oil takes up under the conditions of the above experiment demonstrated that if it be considered as composed of sesquiterpenes, then, for every molecule of the latter it contained two of the gas.

It is evident that cadinene constitutes a large portion of the distillate from Oil of Supa. As to the remaining constituents nothing definite can be said, for no other crystalline derivatives could be obtained from the oil, although many attempts were made. One experiment in this direction was especially interesting because it resulted in the conversion of the oil into a constant-boiling product.

Twenty grams of the distillate were added to 100 cubic centimeters of alcohol and 5 cubic centimeters of dilute sulphuric acid (1 to 5) and heated for seven hours on a water bath with reflux condenser. Afterwards the alcohol was almost entirely distilled and the residue thoroughly shaken with water, after which it was dried and distilled under reduced pressure. The distillate passed over within 5°, leaving a small amount of a tarry residue. On redistillation the product passed over completely at 161° to 163° (37 millimeters). At 760 millimeters it boiled from 271° to 274°. (Cadinene of Wallach, 274° to 275°.) Rotation, +10.2° (10 centimeters, 30°). Specific gravity, $\left(\frac{30°}{30°}\right)$=0.9176. The oil possesses a slight odor which resembles that of cadinene but which is quite distinct from that of the original Oil of Supa. To judge from the boiling point, it would appear to be almost pure cadinene; however, it is dextro-rotatory, whereas the

[1] *Ann. d. Chemie (Liebig)* (1887), **238**, 80.

cadinene prepared from the hydrochloride is strongly laevo-rotatory, and the amount of cadinene hydrochloride which can be obtained from it is no greater than that which can be separated from the original distillate.

The distillate from Oil of Supa absorbs oxygen when it is exposed to the air and it gradually becomes viscous; in a thin layer it slowly hardens; a piece of muslin saturated with the oil and exposed to the action of the air so as to avoid evaporation showed an increase in weight of over 10 per cent in three weeks. This absorption is accompanied by a slight darkening in color. When a current of air is drawn through the oil at 200°, oxygen is very rapidly absorbed and the product becomes viscous and dark colored. The optical rotation of the distillate, removed at ordinary pressure, was found to be only —5.4° (10 cubic centimeters, 30°), while that of the steam-distilled product under the same conditions is—21°. This lowering of the rotation is due to the heating to which the oil is subjected by the former process, it being ascertained that by continued heating at the boiling point, the optical activity is entirely destroyed.

A study of the effect of heat upon the viscosity of the oil demonstrated that very little if any change occurred when it was kept at 250° for ten hours; the specific gravity of the product increased by 0.002, but there was no noticeable alteration in the boiling point.

Non-volatile portion.—This consisted of about 27 per cent of the total and remained as a residue after the distillation of the lower boiling substances at 40 millimeters was completed; after this point, even under greatly diminished pressure, decomposition began. The residue in part consisted of a solid body which separated on the addition of alcohol. It was recrystallized from the latter solvent, filtered to as dry a condition as possible, and then completely desiccated *in vacuo*.

The crystals cling together and are flaky and soft, melting at 63° to 65°. Their boiling point is very high, but heating in a test tube over a free flame causes no decomposition. The body is unaffected by alcoholic potash and it does not evolve hydrogen when treated with melted sodium.

0.2680 gram substance gave 0.8394 gram CO_2 and 0.3602 gram H_2O.
0.1494 gram substance gave 0.4658 gram CO_2 and 0.2010 gram H_2O.

	Found	
	(1)	(2)
C	85.43	85.02
H	14.94	14.61
Total	100.37	99.63

It is evident from the analyses that the body is a hydrocarbon. In chloroform solution it does not add bromine and on warming with dilute potassium permanganate the latter is not decolorized. Substances of the same type have previously been observed in resinous products and essential oils.[2]

[2] Gildemeister und Hoffmann: *Die Aetherischen Oele*, 158.

The saponification number of the residue was found by the usual method to be 64; and under the same conditions the free acid value was 60. These numbers show that the amount of saponifiable matter in the residue is practically *nil*. On taking up the product in ligroin and extracting it with a 10 per cent solution of caustic potash nothing is removed. On digesting it with a small amount of alcohol, which dissolved all but the hydrocarbon already considered, the latter was shown to constitute 25 per cent of the residue and accordingly, about 6 per cent of the original Oil of Supa. The remaining portion, after the evaporation of the alcohol, had the consistency of a thick sirup. On treating this with alcoholic potash on the water bath for several hours it suffered a change and it was then largely soluble in a solution of fixed alkali, notwithstanding the fact that the saponification number indicated no alteration by this treatment.

BALAO: OIL OF APITONG.

Several products from different species of the genus *Dipterocarpus* are utilized by the natives of the Philippines, but in this paper only the discussion of the viscous, slowly drying fluid products which may appropriately be termed wood oils will be entered into. The most widely used are those from the species *grandifluus* and *vernifluus*, the oil from the former being generally known as *Balao*, and the tree from which it is derived as *Apitong;* that from the later as *Malapaho*, the tree being termed *Panao*. Besides the above there are several other wood oils of the same class which are used to a smaller extent. All of these products are similar in composition and consist of a solid resin, of water, and of from 25 to 40 per cent of a volatile oil. From the information which has been obtained it appears that their chief use is in calking small boats and furnishing a protective varnish for wood. For these purposes they are generally mixed with some other solid resin or with lime. At times they are also said to be used for illuminating oils and for torches.

Balao, according to the reports of the Bureau of Forestry, is a product which is in common use in nearly all of the provinces of the Islands. It is secured by allowing the resin to collect in a cup-shaped cavity which is cut in the body of the tree. As the flow decreases, the cup is cleaned, or, if an insufficient supply has passed, the resin is ignited *in situ*, which operation greatly increases the rapidity of the flow, although the product obtained by this means is dark in color, the maximum yield per day from a tree probably being not more than 1 kilo. The freshly exuded resin is white in color, but on standing it soon darkens. When spread upon a surface it slowly hardens to a tough varnish. The samples of this resin which were examined were from three different provinces, there being no doubt as to the botanical identification of the tree. The resin, as it is obtained under ordinary conditions, is a viscous fluid, which is more or less colored according to the time during which it has stood and to the amount of dirt and bark which may have found their way into it. It is not homogeneous, as it contains a large proportion of a granular solid which is immiscible with the fluid, and which remains in suspension. It

possesses a feeble, but characteristic, odor which serves to distinguish it and its oil from other similar products. It appears to dissolve in all the ordinary solvents excepting alcohol, although observations as to solubility are unsatisfactory because of the suspended water which is always present and which does not separate. When the resin is mixed with sesquiterpene oil and the mixture is heated at 140° in an oil bath, some of the water is gradually driven off, although it is necessary to heat to a much higher temperature in order to remove all. A clear solution is finally formed, but on cooling the product, because of the separation of a gelatinous substance, becomes semisolid. Its behavior with fatty oils is precisely the same. A clear solution is formed on heating to expel water, but a precipitation occurs on cooling. This point is important, as it concerns the use of the product for the manufacture of varnishes.

Although Balao is fluid at ordinary temperatures, it hardens when it is treated with steam and it then becomes much too viscous for the latter to penetrate it. Therefore, it is impossible to remove more than a trace of oil by this method. The separation of the volatile products from the resin by distillation under reduced pressure is an impossibility because of an uncontrollable foaming at the beginning of the operation and subsequently on account of the fact that the partially dehydrated residue turns nearly to a solid at the temperature at which the oil passes over. When the resin is gradually heated in a distilling flask which is immersed in an oil bath, only a portion of the water is removed by the time the temperature of the bath has been increased to 200°. In order successfully to remove all of the latter it is necessary to apply a free flame, under which treatment the solid residue gradually melts, and the water, together with a considerable quantity of oil, distill. The continued formation of so much water at a comparatively high temperature, while, at the same time, a large amount of sesquiterpene is being carried over, seems to show that the former liquid is chemically bound in the original oil; however, the combination evidently is not very stable. There is no evidence that the water is formed by destructive distillation resulting from the superheating of the resin, as the distillate is nearly colorless and no odors to be referred to decomposition products are noticeable. Oil continues to distill over on continued heating after all of the water has been driven off; this oil is almost colorless at first, but, as the temperature is increased, the distillate gradually assumes a reddish-green color and at 270° (vapor) there is evidence of decomposition in the flask.

The process just described was the only one found to be practicable for the study of the resin, and accordingly it was applied to the several samples which were examined. In all, about 50 per cent of the original substance was left in the flask as a residue, the remaining 50 per cent consisting of water and oil, the amount of the latter ranging from 22 to 28 per cent. This difference in the oil content may be due to variations

in the manner of gathering the resin, which may permit the evaporation of an indefinite quantity of water, or may be caused by differences in the composition of the resin taken at different times of the year, or from trees of different ages. The residue, after distillation, is a dark-colored, brittle product which evidently is of little value unless it were to yield useful products as a result of destructive distillation. It readily and completely dissolves in chloroform, partially so in ether (leaving a white sediment), not at all in alcohol, and slowly and completely in boiling turpentine. The heating of this resin was continued and it was subjected to a slow destructive distillation, the temperature being so regulated that the thermometer in the vapor did not run higher than 250°. The oil obtained as a distillate constituted 62 per cent of the original by weight and a very small amount of water was formed. On refractionation this product gave 28 per cent below 250° and 40 per cent between 250° and 300°. Beyond 300° the distillate was of the consistency of rosin oil. The first fraction was of a light-green color and contained a very small proportion boiling below 200°.

The oil first obtained by direct distillation of Balao to 270°, after its separation from water, passed over almost completely between 260° and 264° (760 millimeters). No low boiling oil was present. A sample fractioned twice under reduced pressure showed a boiling point of 151° to 154° at 40 millimeters.

Its optical rotation was 78°.5 (10 centimeters, 30°) and its specific gravity $\left(\frac{30°}{30°}\right)$ = 0.9127. It was colored a slight yellow and had the characteristic odor of Balao. Another purified, specimen obtained from a different sample showed the same boiling point. It had a rotation of +87° (10 centimeters, 30°) and a specific gravity of $\left(\frac{30°}{30°}\right)$ 0.9131.

During the removal of this oil from the resin it was necessary to subject the latter to a high temperature by heating with a free flame. Such a process is not favorable to the isolation of a substance in the pure condition, and, as already stated, the oil can not be removed from the resin with steam; so that an expedient was resorted to which consisted of mixing it with equal parts of another non-volatile oil by which means it was possible to isolate the volatile matter from the resin at a lower temperature. Coconut oil which had been thoroughly treated with steam, so as to remove all traces of volatile matter, was used.

The product so obtained redistilled at 149° to 152° (37 millimeters. It was light yellow in color, showed a rotation of +61.3° (10 centimeters, 30°) and a specific gravity of $\left(\frac{30°}{30°}\right)$ 0.9140.

The range in boiling point of this oil is greater than is usually shown by a single, pure chemical substance, but considering its origin and its point of ebullition, it unquestionably is a sesquiterpene, or a mixture of

this class of bodies. Its deportment toward dehydrating agents and toward metallic sodium shows that it contains no substances of an alcoholic nature; furthermore it readily adds halhydric acids and bromine. Despite many attempts, it was not possible to secure from the oil a solid derivative which could be purified by crystallization. Products similar to this one are found in many of the essential oils of commerce, but besides cadinene, which has already been considered in a previous part of this article, in only one case have investigators been able to isolate, either directly or indirectly, an individual substance from such products. By treating certain fractions obtained from oil of cloves and oil of copaiba with a mixture of acetic and dilute sulphuric acid, Wallach[8] isolated a crystalline substance having the empirical-molecular formula required for a sesquiterpene hydrate, the hypothetical mother substance of which was named caryophyllen. The application of this treatment to the oil from Balao did not give rise to a solid derivative. A number of definite bodies, having the general formula $C_{10}H_{16}$ and known as terpenes, have been shown to exist, but it is very seldom that they can be separated in a pure condition by the ordinary physical methods. The application to sesquiterpenes of the methods by which the individual terpenes have been successfully isolated and identified has not led to fruitful results, and a very interesting field is here offered for a more exhaustive study of this interesting and important class of bodies, with a view primarily of identifying and separating the individual components of the sesquiterpene mixtures.

MALAPAHO: OIL OF PANAO.

Several samples of this product, each accompanied by botanical material, were examined. The species was determined to be *Dipterocarpus vernicifluus* Blanco. The resin does not appear to be so widely used as Balao, probably because it dries much more slowly. The method applied to its extraction from the tree is identical with that used for Balao; it is said that a flow of a gallon per day is sometimes obtained. The fresh resin is a white, viscous, sticky fluid having a characteristic odor which serves to distinguish it from other similar products. It absorbs oxygen from the air and on standing becomes dark brown in color. Even when exposed in a very thin film, it hardens very slowly. On heating the resin to 100° its mobility increases, this behavior being different from that of Balao. It appears completely to dissolve in ether and chloroform with the exception of the separation of water; it is only partially soluble in alcohol and benzol; none of its constituents dissolve in water. All of the different samples were found to consist of water, sesquiterpene oil, and solids. On subjecting the resin to distillation with a free flame its behavior is similar to that of Balao. A fresh sample from Ambos Camarines gave 25 cent of water, 35 per cent of oil, and 40 per cent of solid

[8] *Ann. d. Chemie (Liebig)* (1892), 271, 288.

residue. The distillation was discontinued at the point where decomposition became evident. Another sample showed a somewhat different percentage of water and of oil, the difference probably being due to the time and method of collecting.

The sesquiterpene oil obtained from Malapaho redistilled almost completely between 256° and 261° at 760 millimeters, which is a little lower than the boiling point of the oil from Balao. On further purification under reduced pressure a perfectly colorless product was obtained, boiling almost entirely within 3°. Specific gravity, $\left(\frac{30°}{30°}\right)=0.9165$. Rotation, —54° (10 centimeters, 30°).

The solid product resulting from the distillation of Malapaho is similar to that from Apitong, although lighter in color. When subjected to destructive distillation it yields about 50 per cent of a liquid which partly refractions between 200° to 300°, leaving a residue which has the consistency of rosin oil. As was the case with Balao, it was not possible further to identify any of the constituents of the volatile oil of Malapaho.

GENERAL.

The products considered in this article are to be classed with two well-known commercial substances, namely, the balsams of copaiba and gurjun. The Oil of Supa more nearly resembles these balsams than do the other products, since it contains no water and the residue left after distilling the volatile oil is viscous. The *Dipterocarpus* wood oils are composed of a large percentage of water, and after the distillation of the oil, leave solid residues.

Balsam copaiba is a wood oil obtained in South America from various species of the order *Copaifera*. The descriptions found in the literature regarding this product and its volatile oil vary so much and are so contradictory that it is impossible to obtain anything but a very general idea concerning it and its constituents. In general, the balsam may be said to consist of resinous bodies dissolved in a large proportion of a volatile oil which boils between 250° and 275° and may be removed by direct distillation. No definite substances have ever been identified in it or isolated from it excepting the hypothetical sesquiterpene caryophyllen. A sample of copaiba balsam purchased from a drug house in Manila was a dark-colored, transparent, homogeneous liquid. The volatile oil contained in it was removed by distillation under reduced pressure and constituted 38 per cent of the total. There was no water in the product. The oil redistilled within 11° (126° to 137° at 20 millimeters) and was practically colorless. The nonvolatile portion of the copaiba balsam was viscous.

Gurjun balsam (also known as Indian wood oil) is a product widely used throughout southern Asia. Within comparatively recent years it has been introduced into Europe, and is now a regular article of commerce. It is stated to be derived from different species of the *Dipterocarpus* family. The product has never been the subject of thorough

chemical study and the few superficial examinations which have been made and recorded do not show much agreement, other than that the volatile oil boils within a narrow range of temperature at about 255°. The balsam is used in India for preparing varnishes and in adulterating essential oils. It is often stated to be sufficiently mobile to permit of filtration, to possess a fluorescence, and to coagulate on heating. A sample of this product purchased from a drug firm in Manila was found to contain 74 per cent of volatile oil capable of being removed by direct distillation. Only a trace of water was present and the residue was viscous.

The violatile oil redistilled within 2° (154° to 156° at 48 millimeters) and showed the following constants: Rotation, +53° (10 centimeters, 30°). Specific gravity, $\left(\frac{30°}{30°}\right) = 0.9103$.

This gurjun balsam was completely soluble in all the ordinary solvents excepting ligroin, in which it showed a decided turbidity, differing in this respect from the sample of copaiba balsam and also from Oil of Supa; it is also distinct from each of these products in the composition of its volatile oil, as is indicated by the boiling point. Gurjun, as well as copaiba, is produced by making a deep cut, or cavity, in the trunk of the tree near the base. Into this the resin exudes rapidly, amounting in individual instances to from 50 to 180 liters.[4] After the exudation has diminished, a fire is built in the cavity or on the ground below it, which means greatly increases the flow.

From such a general knowledge of the balsams of copaiba and gurjun as it is possible to obtain, it appears that Oil of Supa is a very similar product and that it should prove valuable for the same uses. Copaiba and gurjun, as well as the oil distilled from them, are used for like purposes in medicine. Considering that these oils are not identical and possibly have nothing in common, except that they belong to the class of sesquiterpenes, it seems very probable that the volatile oil from Supa could be put to the same uses. The Oil of Supa could also be utilized in the ways mentioned, namely, in making varnishes, paints, and transparent paper, and in the adulteration of other oils.

In connection with these products the resin from the *Dipterocarpus* tree, Mayapis (Tagalog), is of interest. This product is considered by Dr. Tavera in his book "Medicinal Plants of the Philippines" to be identical with gurjun balsam. A sample of the resin from Mayapis was obtained from Bataan Province. On examination it proved to be quite similar to the two *Dipterocarpus* wood oils which have already been considered. It contained 15 per cent of water and 25 per cent of sesquiterpene oil, which could be removed by careful distillation without decomposition. The residue was quite hard.

[4] Wiesner: *Die Rohstoffe des Pflanzenreiches.*

The oil redistilled at 17 millimeters, possessed the characteristic odor of the resin, and was very light yellow in color. Boiling point, 132° to 140° (17 millimeters. Specific gravity, $\left(\frac{30°}{30°}\right) = 0.9056$.

The *mayapis* resin was light colored, apparently homogeneous in composition, and so viscous that it could scarcely be poured. When heated to 100° it hardened, and exposure to the air produced the same effect, changing it to a pearly, white solid. It dries much more rapidly than do either Balao or Malapaho. The scientific name of the tree yielding Mayapis resin is *Dipterocarpus anisoptera vidaliana,* and Mr. Merrill states that this species has never been encountered outside of the Philippines. Most of the properties of the sample I worked with—namely, the high per cent of water, behavior on heating, low percentage of oil, consistency, and boiling point of the oil—are different from those generally given for *gurjun balsam.*

Balao and Malapaho are largely used by the natives of the Philippines for purposes I have already mentioned, but their application is due to a lack of better material. As varnishes they dry too slowly ever to be considered by the side of modern products. Balao is superior to the other in this respect and gives a very tough and durable coating. The drying properties of these wood oils might be enhanced by the addition of drying oils were it not for the fact that the admixture is not practicable. If practical uses are ever discovered for sesquiterpene oils, then these resins may be of value as a source of the latter, also the products of destructive distillation which have been previously discussed probably largely consist of sesquiterpenes, and at the same time the higher boiling and more viscous portions produced by this decomposition could be put to uses similar to those of rosin oil.

Sesquiterpenes.—Bodies of this type in small quantities have been known for many years, as they occur in nearly all the essential oils of commerce, but the latter generally are expensive and their constituents have been looked upon as rare substances. It is unfortunate and strange that the sesquiterpenes of copaiba and gurjun have never been the subject of serious study. The latter products, as well as those considered in this article, are very largely composed of sesquiterpenes, so that if a demand were ever created for chemicals of this type there would be an abundant supply. The sesquiterpenes would probably be inferior to terpenes for most of the present uses of the latter because of their being practically nonvolatile. However, this property might allow of their use as solvents in various industrial operations where turpentine is inadmissible. Just as is the case with turpentine, they dry slowly in the air, forming a tough varnish, and they might, to a limited degree, profitably be used with the latter for the thinning of paints and varnishes.

In this connection one other point is to be considered. The sesquiterpenes have never received the thorough investigation which they merit

as a distinct and widely distributed class of substances, and it does not seem unreasonable to expect that, when they have received such study, important practical uses will follow as a result. There will then be a demand for the products yielding them. It is a very remarkable fact that we have so high a percentage (from 35 to 80) of volatile sesquiterpenes in most of these wood oils, while the resins yielding terpene oils very seldom have a content of more than 25 per cent of the latter, and the proportion is generally much less.

I wish to return thanks to Dr. H. N. Whitford, of this Bureau, for his valuable and willing help in securing some of the material used in this work.

ORBITOIDES FROM THE BINANGONAN LIMESTONE.

(WITH SOME NOTES ON EARLY CONNECTIONS BETWEEN FORMOSA, THE PHILIPPINES, AND JAVA.[1])

By W. D. SMITH.
(*From the Division of Mines, Bureau of Science.*)

On looking over some samples of fossiliferous limestone collected by Mr. H. M. Ickis, of this Bureau, from the classic Binangonan locality, some forms which resembled *Orbitoides* were noted. Some time later it was possible for the writer to make a trip to this same locality, on which occasion he collected more material and obtained some data with reference to the field relations of the formations.

On closer study, the forms were seen to be, without an exception, species of *Orbitoides,* and no *Nummulites* were detected. However, Richthofen [2] may have seen *Nummulites* there also. Thin sections were made and studied in connection with the admirable sections of similar forms from Formosa and the Riu Kiu Group,[3] which were sent by Professor Koto, of the Imperial University of Tokyo, to Messrs. Newton and Holland and described by them.

In 1862 the late Baron von Richthofen visited a limestone quarry about 4¾ mile northeast of the pueblo of Binangonan on Laguna de Bay, and, according to his account, collected some *Nummulites,* and ever since that date this formation has remained unquestioned, save by Mr. Becker, and referred to the Eocene. So far as we know, Richthofen never figured or described these forms.

[1] This paper the writer intends to serve as an introduction to a field of investigation which he has been assigned to develop as time and opportunity permit. This field, as interesting and important as it is from a scientific point of view, must be made subordinate to the economic work which the writer and his colleagues of this Bureau are at present engaged in. However, it is hoped that articles bearing on this and related subjects will from time to time appear in the numbers of this JOURNAL.

Many statements herein may have to be modified as future work progresses, so that the present conclusions should be regarded more in the light of a working hypothesis than as a definite and final opinion.

[2] *Zeitschr. d. geol. Gesell.* (1862), 14, 357–360.

[3] R. B. Newton and R. Holland: "On Some Fossils from the Islands of Formosa and Riu Kiu," reprinted from *Jr. Coll. Science Imp. Univ. Tokyo* (1902), 17, art. 6.

This is not the first instance of the finding of *Orbitoides* in the Philippines, for in 1901, Mr. Martin,[4] the recognized authority on the little-known paleontology of these Islands, published a short statement concerning *Orbitoides* which were found by Semper in a marl from Alpaco, Cebu.

The importance of *Orbitoides* in the Philippine and Malayan stratigraphy can not be brought out too strongly, for it is a typical zone fossil—i. e., widely distributed, but restricted in vertical range—and from it we have been able to make some interesting and highly important correlations which will be mentioned in the following pages.

FIELD RELATIONS.

Binangonan is situated on the western side of the western of the two peninsulas which extend southward into Laguna de Bay, due east from Cavite. It is reached by launch from Manila by way of the Pasig River.

The surface rock throughout the country immediately north of Laguna de Bay is volcanic and consists of very recent trachytic and basaltic flows, while farther to the north and south is a vast tuff area, familiar to geologists from the literature of Abella, Von Drasche, Semper, Becker, and others. As one goes northeast along the old trail to the limestone, the ground rises rather gradually, until the backbone of the peninsula is reached, at an elevation of about 350 feet, from which altitude the surface drops away in a series of poorly preserved terraces to a broad, flat-bottomed valley on the east.

That this valley was at one time an arm of the Laguna and also of the sea there seems to be little question, for on the highest bench just below the limestone cliff (fig. 1) two shells belonging to the genus *Crassatellites* (marine) were found.

FIG. 1.—Ideal section of part of Binangonan Peninsula.

Almost identical species are living in Philippine waters to-day.

On the western slope of the peninsula also, the evidence of recent uplift is indicated by the deep U-formed stream gorges.

THE VOLCANIC ROCK.

At Binangonan the lava is a dense, bluish-black, clean-cut basalt very much like some phases of the rock from Talim, but a little to the northward in the old city (Manila) quarry, whence the rock was taken for road metal, it becomes lighter colored and more cellular. A section of this has been examined with the petrographic microscope and found to be

[4] K. Martin: "Orbitoides von den Philippinen." *Centralblatt für U. G. P.* (1901), 326, 327.

typical olivine basalt. (Pl. II, fig. 2.) The principal minerals are labradorite, olivine, a green augite, and magnetite. The trachytic texture is very pronounced in the thin section. Zonal structure is very common in the feldspars and an occasional twin in the shape of an X can been seen.

As we travel up the slope to the divide we find the lava becoming more porous and lighter in color, until in the neighborhood of the limestone it is practically a scoria. From the rather limited observations the writer was able to make here, it appears that these flows probably poured out over the country from Talim, leaving a small peak of limestone in part exposed.

THE LIMESTONE.

The limestone, which is the tomb of *Orbitoides,* is exposed in a cliff-like mass, a hundred feet or more in height, and seemingly dipping steeply to the east, though this may prove to be not true bedding, but some secondary structure. In color the rock varies from a light cream to a dirty, bluish-gray. The lighter and denser portions are more fossiliferous. On microscopic examination it was found to contain *Orbitoides,* differing somewhat from the forms described by Messrs. Newton and Holland, and which the writer proposes to call *Orbitoides richthofeni,* some fragments of *Operculina complanata?* Def., and a very imperfect form which is suggestive of *Lithothamnium ramossisimum* Reuss.

DESCRIPTION OF SPECIES.

The genus *Orbitoides* differs in one radical respect from *Nummulites,* namely, in that the chambers of *Orbitoides* are arranged concentrically and not spirally as in the latter form. All the specimens we have found belong to the Lepidocycline group, this terminology referring to the lozenge-shaped chambers along the median plane. It is probable that more than one species is represented, and there is a great difference in the size of some of the specimens.

Orbitoides richthofeni sp. nov.

(Pl. I, fig. 1.)

The type of this is the largest specimen found in this locality, but unfortunately it is not a perfect one. The one we have depicted by Plate I, fig. 1, has lost a portion at each extremity, but if restored would measure in the neighborhood of 36 millimeters in length and 8 millimeters in width at its thickest portion. These tail-like appendages are very characteristic and give to the whole the appearance of the head of a pick.

The initial chamber is not shown, or it is exceedingly small. Instead, along the median plane are developed lozenge-shaped chambers arranged at right angles to the long axis of the form and continued out into the

caudal appendages. The remainder of the chambers are considerably larger and are in certain sections roughly pentangular in outline. Plate I, fig. 2, shows the central portion of the large specimen, much enlarged. In the photomicrograph it is the black band running through the center. Just what the meaning of this is, the writer is unable to determine at the present time, as he has not seen it in a sufficient number of specimens to ascertain whether it is accidental or is some characteristic feature.

Plate I, fig. 2, shows one of the commoner, smaller forms measuring approximately 8 by 4 millimeters. This is unusually circular, but many specimens are almost identical with those figured in Plate 1, fig. 4, of Newton and Holland's paper, and coming from Irometé Island.

We have as yet seen none from the Philippines to correspond to *O. angularis,* figured on Plate I of their paper.

CONCLUSIONS.

As von Richthofen merely mentioned his having discovered *Nummulites* in the Binangonan limestone and never discribed, nor, to our knowledge, figured, any of the species, and as we have not yet found a *Nummulite* from that horizon, we can not find much evidence for calling this formation Eocene.

Furthermore, Orbitoides (*O. verbeeki* Newt. and Holl.), probably the same as our smaller forms (Pl. II, fig. 1), have been found in limestone, in the Riu Kiu Group, which the British paleontologists, Newton and Holland, have placed in the Miocene, and they have been encountered still farther north, in Japan, with *Lithothamnium.* Also Martin[5] has declared the orbitoidal marl of Cebu equivalent to the "Java Gruppe" in which *Vicarya callosa,* the type fossil of the Miocene, was found.

In this connection it is both interesting and due to Becker,[6] who, though he was greatly handicapped in his work at the time of his stay in the Islands by the unsettled state of the country, nevertheless saw enough to make suggestions invaluable to all succeeding workers, to quote him.

> I must confess that the paleontological evidence as to the existence of the Eocene in the Philippines seems to me far from satisfactory. * * * I can see no reason as yet why the Binangonan limestone may not be Oligocene or even Miocene.

Very recently the writer has examined some sections from the Benguet and Lepanto limestones which Mr. Eveland, his colleague, submitted to him, and which lead him to think it quite likely that these beds are the northward extensions of the Binangonan formation. This is not surprising, for we should certainly expect some intermediate occurrences between the Riu Kiu Group and southern Luzon. In certain beds of limestone

[5] *Loc. cit.*

[6] G. F. Becker: "Geology of the Philippine Islands," *21st Ann. Report, U. S. G. S.* (1902), 552.

in the coal field of Batan Island[7] the writer also found *Operculinæ*, although as yet no *Orbitoides*.

The fact that Martin's *Orbitoides* came from a marl, while these we are at present describing occur in a limestone, does not in the slightest degree prevent the inclosing beds from being contemporaneous although they may not be strictly homotaxial.

The bearing of these facts upon the paleogeography, and consequently upon the distribution of the flora and fauna of these Islands, would seem to be exceedingly important. If it can be satisfactorily proved, and these facts appear to contribute something to that end, that the islands of this Archipelago are remnants of a former, more extensive, land mass which was connected with Formosa and Japan to the north and Borneo, Java, and the Malay Peninsula to the southwest, and even with Indo-China and India, much that is now problematical with regard to floral and faunal distribution in this region will have been solved.

This highly interesting problem has been attacked by many naturalists, foremost among whom are R. A. Rolfe[8] and A. R. Wallace.[9] These authors have demonstrated the great and almost confusing mixture of Australian, Indian, Chinese, Formosan, and still more northern types of plants and animals, more particularly the latter, with the endemic forms of the Archipelago. These will not be detailed here, but we shall discuss the distribution and origin of some of these forms.

Mr. Wallace gave two views as to the ancient geography of this Archipelago, one of which, expressed in 1876,[10] maintains that the Islands are truly insular and volcanic and that the union with other Malayan Islands was not of such a nature or duration as to permit of any extended migration on the part of animals. Later, in 1902 in the third edition of Island Life,[11] he gives expression to a second view as follows:

> It is evident that the Philippines once formed part of the great Malayan extension of Asia, but that they were separated considerably earlier than Java and have since been greatly isolated and much broken up by volcanic disturbances; their species have for the most part become modified into distinct local forms, representative species often occurring in the different islands of the group. They have received a few Chinese types by the route already indicated, and a few Australian forms owing to their proximity to the Moluccas. Their comparative poverty in genera and species of the mammalia is perhaps due to the fact that they have been subjected to a great amount of submersion in recent times, greatly reducing their area and causing the extinction of a considerable portion of their fauna.

[7] W. D. Smith: "The Coal Deposits of Batan Island," *Bull. Min. Bur.* (1905), No. 5.

[8] R. A. Rolfe: "On the Flora of the Philippine Islands and Its Probable Derivation." *Jour. of the Linnean Society, Botany* (1884), 21, page?

[9] A. R. Wallace: *Island Life.*

[10] A. R. Wallace: *Geographical Distribution of Animals* (1876), 1, 344.

[11] A. R. Wallace: *Island Life*, 3d edition (1902), 389.

Mr. Rolfe, writing in 1884, favored the former of Wallace's views, but states that "geological evidence will probably in future throw much light on this point."

It appears to the writer that Wallace's later view is more nearly in accordance with the facts. He was familiar at that time with the presence of submerged banks connecting the now isolated groups, but he did not have any paleontological evidence.

Rolfe repeatedly speaks of migrations of southern types from the Malayan and Australian regions northward to the Philippines, but from lack of material he was unable to discuss the great infusion of northern types which must have migrated southward from Siberia, and even North America, through Japan, south China, and Formosa, and which are found in the highlands of northern and central Luzon.

The writer, accompanied by Mr. Merrill, botanist of this Bureau, has recently made the ascent of some high mountains in northern Luzon, where many species of plants identical with, or closely related to, those of Formosa, southern China, and Japan were observed, and also some identical with North American forms, which apparently have migrated from that region by way of the Aleutian Islands, Japan, and Formosa to Luzon.

That there have been in the past repeated land connections between Japan and North America, by the closing of Bering Straits, has substantially been proven by the periodic migrations of molluscan faunas between those regions in past geologic periods.[2] Of course, at the inauguration of the glacial period these plants and animals would migrate far to the south and even into the Tropics. It is expected that future paleontological work will corroborate this view by revealing a decided infusion of Japanese forms in the molluscan fauna of the Pliocene and Pleistocene beds.

If there were such a land connection in Miocene times, as we have already indicated, it is probable that it continued nearly to the time of the present flora, previous to which disruption may have taken place through volcanic disturbances, this break occurring early enough, however, to allow sufficient time to elapse during which the flora and fauna of these Islands could take on their present insular aspect.

We should not fail here to refer to work in a somewhat different field—namely, to the investigations on the distribution of the avifauna in these Islands—made during a number of years by Messrs. Worcester and Bourns.[13] Although their work gives evidence of a great break between the Philippine and Bornean groups, we do not believe their

[12] J. P. Smith: "Periodic Migrations Between the Asiatic and the American Coasts of the Pacific Ocean," *Am. Jr. Science* (1904), **17**, page?

[13] Dean C. Worcester and Frank S. Bourns: "Contributions to Philippine Ornithology." *Proc. U. S. National Museum* (1898), **20**, 549.

conclusions are greatly at variance with our own, for the present distribution of the avifauna might not, and probably did not, go very far back in point of time. Furthermore, birds being far more capable of migration than plants or invertebrate animals, their distribution could not be considered as having as much weight in the evidence as that of the latter. Unfortunately, these investigators did not extend their observations beyond this Archipelago, so that we do not know what their views would have been on this broader problem.

However, it should be stated that the conclusions given above can only be tentative until more is known of the ancient faunas and flora of these Islands and until further study of a comparative nature of the present fauna and flora of China, Japan, Formosa, and the Philippines has been undertaken. It should be stated that Mr. Merrill is now carrying on this work on the flora, and the results of his investigations will be awaited with great interest by all naturalists.

In further support of Wallace's view should be mentioned the occurrence reported, and presumably in the Miocene of Mindanao, of remains of *Elephas* (*Stegodon*) recently identified by Professor Osborn, of the American Museum of Natural History. This *Stegodon* formerly ranged all through southern Asia and is the ancestor of *Elephas indicus*, the living elephant of India.

ILLUSTRATIONS.

(Photomicrographs by Martin.)

PLATE I.

FIG. 1. *Orbitoides richthofeni*, sp. nov.
2. *Orbitoides* sp.?

PLATE II.

FIG. 1. *Orbitoides verbeeki* (?) Newton and Holland.
2. Binangonan basalt.

FIG. 1.

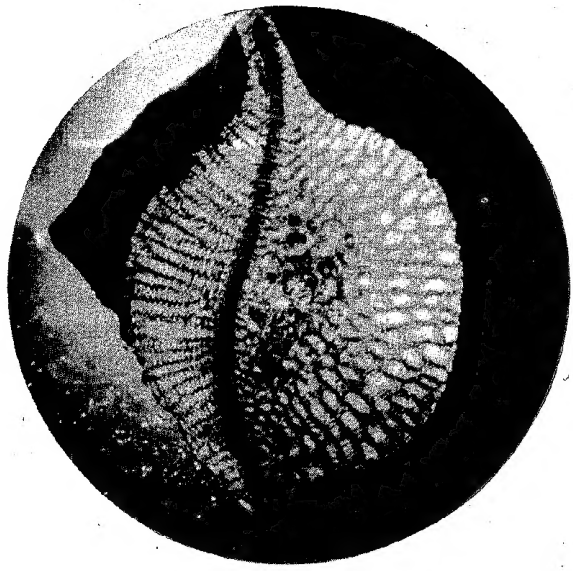


FIG. 2.

PLATE I.

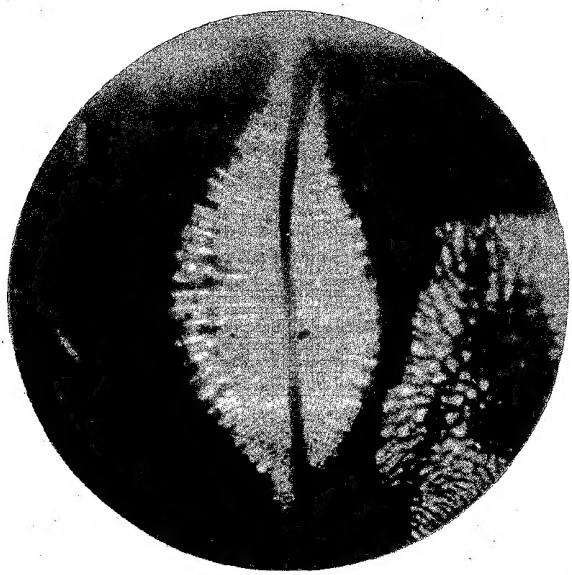


FIG. 1.

FIG. 2.

PLATE II.